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FURTHER STUDIES ON BLOOD GROUPING

II. THE INFLUENCE OF TEMPERATURE UPON ISOHAEMAGGLUTINATION *

By C. G. Guthrie and J. F. Pessel

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For many years it has been recognized that extraneous conditions, and temperature especially, play an important part in immunity reactions such as the agglutination and lysis of bacteria, haemolysis, precipitin formation and complement fixation. That temperature might exercise an equally important influence, however, upon the occurrence of isohaemagglutination has apparently escaped general recognition, despite the fact that attention has been called to this point both directly and by inference.

An interesting and logical introduction to the subject is to be found in the papers of Landsteiner and his collaborators. In their reports [1,2,5] it was shown that, with the cells and sera of various animals, the absorption of normal heterohaemagglutinin was more complete at a lower temperature (6-8° C.) than at a higher (up to 45° C.); that after the union of agglutinin and agglutinogen was formed, it could be broken up by digestion and shaking of the washed, agglutinated cells in salt solution; that this dissociation occurred more completely at higher temperature (45° C.); that the agglutinin thus liberated from the cells could be segregated by centrifugalization and was then capable of agglutinating fresh cells at lower temperature. Observations were also made upon several immune heterohaemagglutinins [6] which were found to behave in a similar manner in their relation to temperature. It was likewise shown that the autoagglutinins [3] found in the blood of various animals were bound at a low temperature (0° C.), but were released and recoverable in relatively pure form at 18° to 20° C. From these investigations, the brilliant work on paroxysmal haemoglobinuria [4,7,8] was a natural development, depending as it did on the application of a similar principle,

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namely, union of haemolysin with erythrocytes in the cold, followed either by dissociation or haemolysis at body temperature, the result being determined by the absence or presence of complement. In their first communication on this subject [4] it was stated that isoagglutination showed a similar relation to thermal influence, as in one case which they had studied the clumping was usually first apparent at a lower temperature at a time when no indication of agglutination was perceptible at body temperature, but it appears that this was not the meaning intended.*

From these observations it is evident that haemagglutinins of several sorts—not only those produced by immunization or those normally present for erythrocytes of alien species, but also those capable of causing agglutination of the red cells of other members of the same species or with the corpuscles of the individual's own blood—are remarkably affected by variations in temperature, in general being much more completely absorbed at low temperature and much more readily dissociated at higher temperature.

In 1911, Ottenberg [9] reported that in absorption tests allowed to stand in the ice-box over night, one volume of cells was able to absorb all the isoagglutinin from sixteen volumes of serum, considerably more agglutinin, in fact, than was required to cause complete agglutination of this amount of cells under the same circumstances.

Koeckert,[10] in 1920, reported experiments in which, after absorption of isoagglutinating sera by susceptible cells at 37° C., partial dissociation and recovery of the liberated agglutinin were effected by digestion of the washed, agglutinated cells in salt solution for one-half hour at 50° C.

The following year Jevrell [11] published more extensive observations on the influence of temperature upon isohaemagglutination but made no allusion to any previous work on this subject. His results were summarized as follows:

> The experiments show a marked difference in the agglutination of red corpuscles at low and at high temperatures.
>
> The most pronounced agglutination is obtained in the ice chest. It is shown that this is due to a quicker and more complete adsorption of agglutinin at low than at high temperature.
>
> After adsorption at 8° the corpuscles again lose part of the agglutinin when brought into higher temperatures.
>
> The experiments seem to indicate the maximal adsorption of agglutinin is different at different temperatures and more nearly complete at the low than at the high temperatures. When, therefore, the adsorption has been carried out at a low temperature and the corpuscles after that are placed at a higher temperature, they can retain only the quantity of agglutinin that corresponds to the maximum for the respective temperature and accordingly lose agglutinin until this maximum is reached.

Important and comprehensive as were these observations beginning with those of Landsteiner, there is an interesting aspect of the influence of temperature upon isohaemagglutination which has been left untouched.

Object

It has been shown that an agglutinating serum acts on erythrocytes more rapidly and more effectively at a low than at a high temperature. Although the titre of any isoagglutinating serum would thus probably be less when tested at 37° C. than at ice-box temperature, a powerful serum, when undiluted, would doubtless give a positive result with susceptible cells at both temperatures. But what would be the result with a weaker serum under similar conditions? Might not an undiluted serum contain so little agglutinin as to cause no clumping except at a lower temperature? Would not the presence of isoagglutinin in such a serum escape recognition altogether when tested by the ordinary methods?

These questions seem to have a direct bearing upon the detection of incompatibilities in blood-matching tests preliminary to transfusion; if so, they are of practical importance. The present communication, which is the record of an attempt to supply the answers, was stimulated by the following experience.

Personal Observations

In the course of some experiments to be reported at another time, cross-agglutination tests were made with 10 different bloods, all of which, by the usual standards, belonged to the same group. Each of these tests was examined four times, as follows: (1) immediately, to see whether the preparations were satisfactory; (2) after one hour at 37° C.; (3) after two hours at 37° C.; (4) after standing overnight at room temperature. One test in this series of 100 (sera *vs.* r. b. c. from 10 persons) was negative at the first three examinations but positive at the fourth. In other words, this test (ser. C. E. W. *vs.* r. b. c. P. O. C.) was negative when first set up, was still negative after one hour and two hours in the incubator, but had become positive on standing over night at room temperature. All of these tests had been set up in duplicate and it was interesting to note that this pair showed the only positive results in the entire series.

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* The statement referred to was as follows: "In einem untersuchten Falle bei den Isoagglutininen, tritt dieses Verhalten dadurch besonders auffällig hervor, dass die Agglutination überhaupt erst bei niederer Temperatur deutlich wird, während bei Körpertemperatur Agglutinationserscheinungen nicht wahrzunehmen sind."

In a personal communication from Dr. Landsteiner we were informed (1) that the sentence quoted above contained a misprint as originally published and that for *Isoagglutininen* one should read *Autoagglutininen*; (2) that he was familiar with the fact that the union of isoagglutinin with isoagglutinogen may be dissociated by heating; but (3) that he had published no work on this subject.

As the chief elements in which the fourth examination differed from the preceding ones were time, light and temperature, the influence of each in the production of this agglutination was accordingly tested with the same serum and cells as those used in the first preparations. The two original tests were placed in the incubator and at the end of two hours had become completely negative, but agglutination was again very marked after the same length of time at room temperature. As this experiment was repeated several times with the same preparations, it was concluded that either light or temperature rather than time, was probably the deciding factor. To clear up these points several new preparations were made; four of these were placed in the dark at 37° C.; four in the light and four in the dark at room temperature (25° C.); and four in the ice-box at 12.5° C. When examined at the end of 15 minutes, the result was strongly positive in the preparations placed in the ice-box; later it became positive in those kept at room temperature but no difference, either in rapidity or extent of agglutination, was discernible between the preparations exposed to, and those protected from, the light; there was still no agglutination after 24 hours in those placed in the incubator.

These results were so convincing as to the effect of temperature on these particular specimens of blood that it was obviously desirable to repeat the tests with other specimens from the same persons. This was done on two occasions in a period of two weeks, and several dozens of tests were made with the serum of C. E. W. and cells of P. O. C. In all of them the effect of temperature was very pronounced,—strong agglutination in the ice-box; no agglutination in the incubator; and at room temperature a wide range of variation, extending from strong agglutination to none whatever, depending on the actual temperature of the room at the time of the test. Thus it was found that agglutination was marked at 15 to 20° C. or below; definite at 20 to 25° C.; moderate to slight up to 30° C.; and absent entirely above 32° C. Under the circumstances, the question naturally arises as to whether we were dealing with an iso- or an autoagglutinin reaction.

As has been stated, the two bloods in question (C. E. W. and P. O. C.) belonged to the same one of the four accepted groups. This was determined in the first place, by the reaction of cells and serum against the serum and cells of one member of "Group II." It was then tested for each blood by cross-agglutination with the blood of a member of each of the four accepted groups. The blood of C. E. W. and that of P. O. C. behaved exactly alike in all these tests. They were likewise tested by cross-agglutination against the blood of 18 other persons, each of whom was, by the usual standards, a member of their own group. At 37° C. there was no agglutination; at ice-box temperature the only positive test was the one previously found (ser. C. E. W. *vs.* r. b. c. P. O. C.). The criteria for the recognition of an autoagglutinin [12] are that it is capable of agglutinating: (1) the corpuscles of the individual in whose serum it occurs; (2) the corpuscles of any member of the same group; (3) the corpuscles of the other groups; (4) any of these various types of corpuscles only at low temperature, and at body temperature is entirely dissociated from its union with the red blood cells. It is true that the agglutinin in question exhibited the temperature relations just mentioned, but this has been shown to be a property which is shared to a considerable extent by haemagglutinins of other types as well. Inasmuch as the peculiar haemagglutinin in the serum of C. E. W., however, was incapable of clumping the cells of C. E. W., in other words, since it did not cause autoagglutination,—further proof is unnecessary; it was not an autoagglutinin. Moreover, it did not agglutinate the cells of 18 other members of the same accepted group; it was always and only operative against the cells of a particular member of this group (P. O. C.). The relations of each of these two bloods (C. E. W. and P. O. C.) to the other accepted groups were those of the group to which the usual tests assigned them. Absorption experiments, to be reported in detail at another time, showed that the isoagglutinins present in the sera of these two bloods could be absorbed separately, leaving only the one capable of agglutinating the cells of P. O. C., or removing this and leaving those operative against the other accepted isoagglutinin groups. The cross-agglutination and absorption experiments were repeated so many times as to leave us in no doubt regarding the agglutinin-agglutinogen content of these two bloods. We have, therefore, no hesitation in asserting that the agglutination of the cells of P. O. C. produced by the serum of C. E. W. was caused by the union of a specific isoagglutinin with its corresponding isoagglutinogen.

Titration of the serum of C. E. W. showed that in a dilution of 1 to 16 it was capable of causing agglutination of the cells of P. O. C. in the ice-box (15° C.), but that no clumping occurred at 37° C. even with undiluted serum. We therefore endeavored to ascertain whether a comparable difference in titre at these two temperatures could be demonstrated in a strong isoagglutinating serum. The serum of a "Group IV" blood (P. J.) having the formula *ABC—o* and two types of "Group II" cells (J. F. P. and C. G.) one containing isoagglutinogen *b* and the other both *b* and *c*, were used for this purpose. The result of the titrations are shown in Table I.

From the results shown in Table I it is apparent that:

(1). Agglutination occurred in higher dilution of serum at ice-box (15-16° C.), than at room temperature (23-24° C.); and in higher dilutions at room, than at incubator temperature (36-37° C.)

(2). In diluted serum the time element plays an important part, since with both types of "Group II" corpuscles, at any of the three temperatures used, agglutina-

TABLE I.

Titrations of isoagglutinating serum at different temperatures.

Serum	R. B. C.	Temperature	Time	AGGLUTINATION BY VARIOUS DILUTIONS OF SERUM											
				1:1	1:2	1:4	1:8	1:16	1:24	1:32	1:40	1:48	1:56	1:64	1:72
Group IV (P. J.)	Group II (C. G.)	36°-37° C.	1 hr.	5	4-5	4	2-3	±	0	0	0	0	0	0	0
Group IV (P. J.)	Group II (C. G.)	23°-24° C.	1 hr.	5	4-5	4-5	3-4	1-2	±	0	0	0	0	0	0
Group IV (P. J.)	Group II (C. G.)	15°-16° C.	1 hr.	5	5	5	4-5	3-4	1-2	±	0	0	0	0	0
Group IV (P. J.)	Group II (C. G.)	36°-37° C.	2 hrs.	5	4-5	4-5	3-4	1-2	1	±	0	0	0	0	0
Group IV (P. J.)	Group II (C. G.)	23°-24° C.	2 hrs.	5	5	5	4-5	2-3	2	1-2	1-2	1-2	1	±	0
Group IV (P. J.)	Group II (C. G.)	15°-16° C.	2 hrs.	5	5	5	5	4-5	3-4	3	2	1-2	1-2	1-2	1
Group IV (P. J.)	Group II (J. P.)	36°-37° C.	1 hr.	4-5	3	1-2	±	0	0	0	0	0	0	0	0
Group IV (P. J.)	Group II (J. P.)	23°-24° C.	1 hr.	4-5	4-5	3-4	2	±	0	0	0	0	0	0	0
Group IV (P. J.)	Group II (J. P.)	15°-16° C.	1 hr.	5	4-5	4	2-3	1-2	±	0	0	0	0	0	0
Group IV (P. J.)	Group II (J. P.)	36°-37° C.	2 hrs.	5	3-4	2-3	1	±	0	0	0	0	0	0	0
Group IV (P. J.)	Group II (J. P.)	23°-24° C.	2 hrs.	5	5	4-5	3-4	1-2	±	0	0	0	0	0	0
Group IV (P. J.)	Group II (J. P.)	15°-16° C.	2 hrs.	5	5	4-5	4-5	3-4	1-2	1-2	1	±	0	0	0

The numbers in the fifth and succeeding columns represent the degree of agglutination, 5 being used to indicate maximal clumping and ± a questionable result.

tion occurred in much higher dilution after two hours than after one hour.

(3). The difference in agglutinative strength at these three temperatures was pronounced; for the cells of C. G. the titre was 8, 16 and 24 at one hour, but 24, 56 and 72 at two hours; for the cells of J. F. P. the titre was 4, 8, 16 at one hour but 8, 16 and 40 at two hours.

These titrations serve to illustrate how a relatively weak agglutinin could cause manifest clumping in the cold and escape recognition entirely at incubator, or even at room temperature. They also render intelligible our observation that the agglutinative titre in the serum of C. E. W. for the cells of P. O. C. was 16 at 15° C. and 0 at 37° C.

Comment

The observations of Landsteiner and others make it apparent that in the clumping of erythrocytes by agglutinins of several kinds, temperature is an important factor,—often the deciding factor. The work of Jevrell emphasizes the relation of temperature to isohaemagglutination in particular. Our experience shows that the presence of isoagglutinins cannot always, nor their absence ever, be determined with assurance unless the temperature factor is considered.

Some workers are accustomed to read the results of isoagglutination tests after the preparations have remained for a time in the incubator, on the theory that conditions are thereby secured in which the temperature is not only constant but approximately that which obtains *in vivo*. This was formerly our custom, but we now realize that 37° C. is not the most favorable temperature for such reactions. The great majority of workers, however, make their readings on preparations which have stood for a longer or shorter period at "room temperature." From this one might, perhaps, infer the temperature of a well regulated room,—70° F. or 21.1° C.; actually it may be something quite different. In our laboratory, the room temperature from May 5 to June 20, 1923, varied between 18.5° C. and 37.5° C. Under such circumstances, an unqualified statement that an experiment has been conducted at room temperature, is only comparable to the use of that term, well known in mensuration, "about as long as a piece of string."

Ottenberg[9] noted that in tests made with ascending dilutions of an isoagglutinating serum, the agglutinated clumps of cells became smaller and smaller as the limit of potency was approached; this observation we have had many opportunities to confirm. Unger,[13] in commenting on the behavior of what he calls "minor" agglutinins as distinguished from the "major" agglutinins responsible for the agglutinative propensities of the accepted blood groups, said:

> In the course of performing many thousands of tests preliminary to transfusion, I have occasionally noted that although the donor and patient are of the same group, when the bloods are tested one against the other a small number of agglutinated clumps of blood cells will be seen.
>
> Although ordinarily such donors are rejected, urgent need has forced me to transfuse patients in which this phenomenon occurred. In each case there was a sharp reaction—chill and fever—undoubtedly due to the occurrence of agglutination.

In view of our experience, it is but natural to speculate concerning the degree of clumping which would be produced by a "minor" agglutinin permitted to act on susceptible cells at an optimal temperature.

That two isoagglutinating sera may differ from each other in titre has long been recognized, but it is now evident that the demonstrable strength of any such serum

is markedly dependent upon the temperature at which the titration is performed; or transposed to the interrogative form, if the same serum has a different strength at different temperatures, how can we compare the strength of two sera unless they are titrated at the same temperature?

A strong serum may cause agglutination both at 37° C. and at ice-box temperature, but it occurs earlier and more completely at a lower temperature; in other words, more agglutinin is required to produce clumping at body temperature than in the ice-box. On the other hand, a weak serum, which when tested 1:1 gives a positive result only at a lower temperature, may cause agglutination even at 37° C., if enough serum is used.

To our suggestion that isoagglutination tests should be performed in the cold, it may be objected that the presence of autoagglutination might lead to confusion. It may be replied (1) that autoagglutination has been very rarely observed in human blood. (2) Control tests in which the serum of each blood is tested against its own cells, have always formed a part of our technic; they promptly reveal autoagglutination in the cold. (3) If such control tests were generally employed, the presence of an autoagglutinin might be detected more often and so cause less confusion than at present when it is apt to pass unrecognized or to be mistaken for a true isoagglutinin from its ability to act upon cells of any group at some room temperatures. (4) A serum shown to contain autoagglutinin may be freed from it by absorption with its own cells at 15° C. or below; interference from this source thus removed, one may proceed with tests for isoagglutination.

If it merely requires more agglutinin to give a positive test at a higher temperature, why not use more serum in proportion to the number of cells, instead of lowering the temperature? The question is reasonable; so, we believe, is the answer. The agglutinative strength of the serum of C. E. W. for the cells of P. O. C. was 16 at 15° C. and 0 at 37° C., as determined microscopically in preparations containing one drop of serum and a like amount of a very weak suspension of cells. It required 12 drops of serum to one drop of the same cell suspension to render the faintest degree of agglutination perceptible at 37° C., and 16 drops to make it distinct. This makes an unwieldy bulk for hanging-drop preparations, necessitating preliminary mixing on a slide or in a test-tube and subsequent transfer of a portion to the coverslip. Even then, the proportions used render the number of cells in the preparation too few for satisfactory results. This objection applies equally when the volume of serum is kept constant and the suspension of cells diluted with salt solution. In either case, the preparations are less satisfactory, the results less clear-cut and the simplification in the technic *nil*.

The relation of temperature to isohaemagglutination reactions is not a matter of merely academic interest. Failure to take it into consideration may readily explain the occasional discrepancies in results obtained with the same blood at different times, many of which have been reported but attributed to a variety of other causes such as drugs, diet, disease or spontaneous fluctuation in agglutinative power. In like manner, we may perhaps interpret the varied classification or grouping of the same blood in different laboratories; instances of this sort are not uncommon. Chiefly, however, we may thus account for certain unexplained post-transfusion reactions, through failure to recognize incompatibilities which would have been evident in tests performed at a more favorable temperature. It is, no doubt, superfluous to indicate that under these circumstances, the recipient would not need to be cooled to this optimal temperature in order to agglutinate the transfused cells; if the agglutinin in his serum were present in sufficient volume, agglutination could be accomplished—more slowly, less dramatically but quite effectively,—at the temperature of his body.

As the serum of C. E. W. contained a specific isoagglutinin for the cells of P. O. C., it is obvious that these two bloods differed from each other and from the 18 bloods incapable of agglutinating the cells of P. O. C. or of being agglutinated by the serum of C. E. W. That all of these 20 bloods should be assigned to the same group, is an interesting commentary on the accepted methods of classification.

Conclusions

1. Temperature is an important factor in isohaemagglutinin reactions.

2. Agglutinative titre is higher and agglutination occurs more rapidly at ice-box than at body temperature.

3. More isoagglutinin is required to cause clumping at body than at ice-box temperature. Thus an amount of agglutinin sufficient to cause marked agglutination in the ice-box may escape recognition in tests made at 37° C. or at room temperature.

4. Isohaemagglutination tests should be carried out at a *constant* temperature. For this purpose, a temperature of 15° C. is better than body temperature and greatly to be preferred to the wide thermal range indicated by the term "room temperature."

5. Time is also a factor to be considered in agglutination by diluted sera. It is probably of similar importance in tests with weak undiluted sera. In our experience readings made on preparations that have stood for one and two hours in the ice-box are within the limit of safety.

6. The possibility of confusion from the occurrence of autoagglutination is remote; it may be easily controlled.

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AN ANALYSIS OF THE CLINICAL HISTORIES OF PATIENTS WITH PERNICIOUS ANEMIA IN THE JOHNS HOPKINS HOSPITAL FROM 1918 TO 1922 INCLUSIVE

By Charles R. Willson and Frank A. Evans

Pernicious anemia is a term which should not be used to designate an anemia of unknown etiology, nor to indicate merely a certain type of anemia frequently encountered in many and various conditions. It should be restricted to a definite clinical syndrome, and its use should bring to mind not only an anemia of unexplained cause or a certain blood picture, but a complex disease having many characteristic features. The manifestations of pernicious anemia vary considerably; and many conditions may be encountered which strongly resemble it, conditions in which a cause for the anemia can be demonstrated and which are based on etiological factors probably entirely different from those operative in pernicious anemia. The diagnosis of pernicious anemia does not depend upon one feature alone, or on several features. Every detail of the clinical picture should be sharply scrutinized if error is to be avoided. Many good clinical descriptions of the disease are available, but in none are the smaller details of the clinical picture discussed closely enough, or attributed enough significance in diagnosis.

This paper gives the results of an analysis of the details of the clinical histories of 111 patients suffering from pernicious anemia—so diagnosed and treated in the Johns Hopkins Hospital from 1918 to 1922 inclusive. Only those histories are included from which, upon final analysis, the diagnosis of pernicious anemia was beyond question, according to the strictest criteria. Thirty of these patients lived in Maryland, and the rest came from various parts of the United States, no one state or section predominating.

Race.—Of the 111 patients only eight were negroes, two women, and the rest men. All were mulattoes. There were not as many beds in the hospital available for negroes as for white people, and the admissions of negroes are therefore somewhat less. But this fact does not suffice to account for the rarity with which a diagnosis of pernicious anemia was made in colored people, and seems to warrant the conclusion that the disease is rare in this race. Again, it might be interesting to note here that seven of the eight negro patients had very complete remissions in the hospital, and gave other evidences of being much less severely affected than the majority of the white patients.

Sex.—Of our 111 patients 82 were men and 29 women. From this it would seem that the disease is more common in men than women; but the series is too small to permit reliable conclusions. According to the earlier German clinicians the disease is much more common in women than in men. This, however, has not been confirmed by later investigators, the Germans being probably led into error because they did not draw the lines of diagnosis so sharply, and included many cases of women suffering from an anemia associated with various phases of child-bearing. Osler in his textbook expresses the opinion that the disease is more common among men. In a series of 143 cases studied by Levine and Ladd [1] in Boston, there were 59 men and 84 women. If now these figures are combined with ours, in a total of 254 patients there were 141 men and 113 women. Stengel [2] also found the disease slightly more frequent in men than in women. Examination of numerous statistics, however, would lead to the conclusion that the incidence of true pernicious anemia is about the same in men and women, or only slightly more frequent in men.

Age.—Our patients were distributed according to age in different decades as follows (Table I.):

TABLE I.

Years	No. of cases	Per cent of total
0—10	0	0
10—20	1	.9
20—30	3	2.7
30—40	17	15.
40—50	26	23.
50—60	37	33.
60—70	18	16.
70—80	9	8.
80—90	0	0

Pernicious anemia is recognized as a disease of adult life and these figures indicate the correctness of this conception. Thus, between the ages of 40 and 60 years 56% of our cases are found, and between 30 and 60 years, 71%.

Although the disease occurs in old age, it is much less common before than after 60 years. At the other extreme of life, however, it is very rare, for we had but one patient under 20 years of age and none under 10. Dr. Howland states that among the 36,000 non-surgical patients under 14 years of age treated in the Harriet Lane Home from its opening until 1922, not one case of pernicious anemia has been encountered, and adds that he has never seen a case of pernicious anemia in a child, thus enlarging greatly on the thirty-six thousand patients seen by him in the hospital.[3] In view of this vast number of carefully studied cases which did not include one patient under 14 years of age with pernicious anemia, one may justly be inclined to question the reported cases. Various kinds of splenic anemia are, of course, common in children, and occasionally a case is encountered which is difficult at first to differentiate from pernicious anemia.[4] Furthermore, in children unimportant disturbances may give rise to bizarre changes in the blood of no great moment, but which, if encountered in an adult, would be strongly significant. Such blood changes might lead to confusion, and any case in the literature reported on the basis of the blood findings alone is open to serious question.

Family History.—The family histories from the general run of ward patients in a large hospital do not inspire confidence in their accuracy, and one hesitates to draw conclusions from them. This is especially true in regard to the occurrence in the family of pernicious anemia, the critical diagnosis of which is sometimes difficult even under the best circumstances, and probably quite impossible for physicians with large general practices and without hospital or laboratory facilities. For this reason our patients were merely asked whether any members of their families had had chronic anemia, and no serious attempt was made to determine whether or not the anemia mentioned was pernicious anemia. In our series three brothers of one patient, one brother of two others, one sister of another, and the mother of one had died with some form of anemia. The mother of the last patient also had a cancer, so that the anemia here can readily be explained. There were, including the last patient, only five among the total 111 patients who had other members of their families die from anemia. This percentage (4.5) is not striking. In the series of patients reported by Levine and Ladd, however, 6.3% gave histories of pernicious anemia in the family. If these were, as the authors believed them to be, examples of true pernicious anemia it certainly points to a familial occurrence of the disease.

Tuberculosis was reported in the families of fourteen (12%) of our patients. Two of these patients had more than one relative who had suffered from tuberculosis and twelve had only one. In the families of sixteen (14.4%) cancer is recorded, one member being affected in fourteen, and two members in two. Diseases of the kidney caused death in families of eight (7.2%) of our cases. Heart trouble was mentioned in the family history of thirty-five patients (31.5%).

Fifty-one of our patients gave family histories that did not include deaths from anemia, tuberculosis, cancer, kidney or heart trouble, and in which there was no record of metabolic or nervous disorders, or any blood disease. This suggests that, if the histories can be relied upon, pernicious anemia occurs with just as much frequency in people coming from families in which there is no taint, as among those less fortunate.

Occupation, Habits, and Previous Illnesses.—The occupations recorded in the histories of our cases include common laborers, stevedores, miners, carpenters, farmers, clerks, merchants, army officers, milliners, teachers, housewives, lawyers, ministers, physicians, ladies of leisure and retired capitalists. Just about as many and different occupations are mentioned as could reasonably be expected among any 111 people chosen at random. It would seem, then, that occupation is not a factor in the causation of the disease and that it is just as likely to occur among those enjoying the better things of life as among others less fortunate who have little or no ease or luxury. Similarly, the histories reveal no dissipation, or habit of eating, drinking, sleeping, working, sexual activity, or manner of life that would seem to have any importance in relation to the present illness.

Among the previous illnesses, however, it was noticeable how frequently typhoid fever, pneumonia, and malaria were recorded. Thus, of the 111 patients, twenty-four had had typhoid fever, sixteen had had pneumonia, and ten had had malaria. Four patients had had typhoid fever and pneumonia, five typhoid fever and malaria, and one patient had had all three. No other illnesses were recorded frequently enough to attract attention. It was

noteworthy that none of the patients had had any cause for chronic blood loss or blood destruction.

Complaint.—By far the most common complaint was weakness, this being given by sixty-six (59%) patients. The next most common was some symptom pointing to the gastro-intestinal tract, which occurred in thirty-two (29%). These were distributed as follows (Table II):

TABLE II.

Complaint	No. of cases	Per cent of total
Stomach trouble, dyspepsia, and indigestion	12	11
Diarrhoea	9	8
Nausea and vomiting	5	5
Anorexia	2	2
Sore mouth	2	2
Biliousness	1	.9
Abdominal discomfort	1	.9

Tingling in the hands and feet, numbness of the hands or feet, or ataxia was mentioned by twenty-seven patients (24%). Eighteen patients (16%) complained of anemia, and among these, two were specific and mentioned pernicious anemia. Next in order of frequency comes dyspnoea which was mentioned by eleven of our patients (10%). Five of our patients (4.5%) complained of swelling of the feet and legs. Four (3.6%) complained of nervousness; four (3.6%) of pain in back, hips, and legs; three (2.7%) of loss of weight; two (1.8%) of vertigo, and two of mental deterioration. The following symptoms were mentioned once: cough, headache, rheumatism, buzzing in the ears, a general breakdown, and general pains.

Only ten patients did not mention at least one of the three most common symptoms, weakness, some disorder of digestion, or sensory disturbances of the extremities. These ten patients included four who complained of anemia alone and two who knew they had pernicious anemia, their complaint being probably a repetition of a diagnosis they had been told elsewhere rather than a personal record of symptoms. If these patients are excluded, among the remaining 105 there were only four (3.8%) who did not lay stress upon one of the three most common symptoms mentioned above. These patients complained of mental deterioration, vertigo, leakage of the heart, and pain in the right hip. The man who complained of his heart really had chronic valvular disease and, having been told of this diagnosis, no doubt attributed all his symptoms to this condition. Again, the patient whose only complaint was pain in the right hip did have, among his other troubles, disease of his sacro-iliac joint. Eight of these ten patients, when asked their first symptom, said it was weakness, and the remaining two mentioned at least one of these symptoms at some time during their stay in the hospital. Thus, there were none of our patients who did not recognize at one time or another as prominent among their disturbances either weakness, indigestion, or sensory abnormalities of the extremities.

Symptoms.—The first symptoms attracting the attention of the patients were very much the same as their complaints. The list is as follows (Table III):

TABLE III.

Symptoms	No. of cases	Per cent of total
Weakness, or easy fatiguability	46	41.4
Nausea, vomiting, diarrhoea, indigestion	19	17.1
Constipation	1	.9
Sore tongue	8	7.2
Sensory disturbances of extremities	19	17.1
Oedema of legs	5	4.5
Loss of weight	2	1.8
Dyspnœa	6	5.4
Yellow color	2	1.8
Nervousness	2	1.8
Pallor, pains in back, hallucinations, headache, dizziness, sleeplessness, hematemesis, salivation.— Of each	1	.9

This list of first symptoms practically covers those complained of by the patients while in the hospital. Because several of them were frequently given by the same patient when asked to mention all his troubles, the frequency of occurrence of many of them is increased in the study of the complete history. Thus, weakness and fatiguability are recorded at one time or another in the histories of eighty-seven of our patients (78.4%). Twenty-three patients (26.7%) complained of sore tongue. In sixty-five (58.6%) gastro-intestinal symptoms were very outstanding during the early days of their stay on the hospital. Of these the most common were anorexia, nausea, vomiting and diarrhoea. Symptoms and signs pointing to disturbances of the nervous system were present in eighty-five (76.6%) of the patients.

Duration of the disease.—Attempts to determine the duration of the disease in our patients was so unsatisfactory that nothing of value can be deduced from them. Two patients said their first symptoms appeared two months before coming to the hospital; one patient insisted he had had the disease seventeen years. One patient claimed to have been sick nine years, four for seven years, one for six years, and six for five years. The statements of all the other patients on this point lay between these extremes. Many patients, of course, dated the onset from an attack of influenza during 1917 and 1918.

Physical Examination.—In discussing the physical findings it will be impossible to give in full all the notes on these patients, or indeed to mention many of them. Accordingly, only the more pertinent points will be pre-

sented in tabular form—only those findings that are very frequent, or of especial importance.

None of these patients were emaciated or markedly run down. A note that the state of nutrition was poor occurs in the histories of forty-four patients (39.6%). Only six (5.4%) of these stated that they had lost weight and gave evidence of it; but in none did it appear that any recent loss had been very severe. All the rest of the patients, sixty-seven (66.3%) were well nourished and one patient might be considered obese, being about 15 pounds over his ideal weight. This last patient was not, it is true, in the last stages of the disease when the study of his case in the hospital was made. On entrance he had 2,012,000 red blood cells and 45% of hemoglobin, and when discharged 22 days later after four transfusions of citrated blood he had 4,070,000 red blood cells and 83% of hemoglobin. The state of nutrition, however, bore no relation to the severity of the disease, patients with a severe anemia and apparently quite ill often looking well nourished. Indeed, such an observation is a very helpful diagnostic point.

The *color of the skin* in three (2.7%) of our patients is recorded as good. One of these patients came into the hospital with over 3,000,000 red blood cells, 80% of hemoglobin, 7,400 white blood cells and 73% of polymorphonuclears. The other two had a lower red blood cell count on admission, but never a severe leucopenia nor a diminished percentage of polymorphonuclear cells. All made prompt recoveries, two of them without transfusion and the other with only one. They were, therefore, in the earlier stages of the disease. In all the other patients, 108 (97.3%), the skin is noted as being discolored. Different terms are used to describe the color—pale yellow with brown tint, lemon yellow, sallow, ashen, icteroid tint, pale and slightly yellow.

Condition of Teeth.—Sixteen (14.4%) of our patients had had all their teeth extracted. Twenty-eight (25.2%) had good teeth. In all the remaining sixty-seven (60.4%), the teeth were in bad condition.

Such statistics do not mean much in a report based on patients the majority of whom were ward cases. If any conclusions may be drawn from them, it is that pernicious anemia and good teeth are often seen together. Sixty-five (58%) of our patients were free from pyorrhoea and had healthy gums. Only one patient had bleeding gums. Forty-five (39.6%) had pyorrhoea, but in twenty-one (18.9%) of these it was slight.

Papillary atrophy of the tongue was marked in only forty-three (38.7%) of our patients, and moderate in seven (6.3%). In forty-six (41.4%), it was noted as absent. In the remaining fifteen patients papillary atrophy was recorded as very slight by some observers and as absent by others. Although papillary atrophy was not a very constant finding, *a clean tongue was always present.* A coated tongue and a diagnosis of pernicious anemia is a combination to excite suspicion.

The *thyroid gland* in eighty-four (75.6%) of our patients was normal. The isthmus was slightly enlarged in ten (9%). There was a general enlargement, implicating both lobes in fourteen (12.6%) of the patients, and nodules in the isthmus of three. Other than easy fatigability, none of these patients showed any signs of hyperthyroidism, there being no exophthalmos, tachycardia, tremor, or emotional instability noted.

The *peripheral arteries* were normal in fifty-five (49.5%) of our patients. The remaining 50% showed various grades of arteriosclerosis, only nine (8.1%), however, exhibiting anything more than very moderate thickening. The number of times the arteries were palpable differed in no way in relation to age, occupation and habits, from what might be expected in any series of patients. The same may be said of the blood pressure. Nine patients (8.1%) had systolic blood pressures under 100 and in seventeen (15.3%) it was over 140. In the remaining eighty-five (77%), the systolic pressure was between 100 and 140. The systolic blood pressures, when tabulated, appear as follows (Table IV):

TABLE IV.

Systolic blood pressure	No. of cases	Per cent of total
70— 80	1	.9
80— 90	1	.9
90—100	7	6.3
100—110	23	20.7
110—120	23	20.7
120—130	17	15.3
130—140	10	9.0
140—150	11	9.9
150—160	3	2.7
160—170	2	1.8
170—180	1	.9

The difference between the systolic and diastolic blood pressure as found in these cases showed nothing characteristic. A few of the patients had a relatively low pulse pressure, and about the same number a rather high one. Thus, it was under 40 in sixteen (14.4%) of the patients, and over 70 in fifteen (13.5%). In the remaining eighty cases it was between 40 and 70.

Analysis of the records for *enlargement of the spleen* are illuminating. Notes that the spleen was not palpable occur in the histories of eighty-nine (80%) of the patients. In many of these there is the additional observation that it was not enlarged to percussion; and in six (5.4%) increased splenic dullness was recorded. One patient could not relax his abdomen sufficiently to permit a satisfactory examination. In the remaining twenty-one (19%) the spleen was palpable; in one it was 2 cm., in four, 3 cm., in two, 5 cm., and in one it was recorded as a hand's

breadth below the costal margin. In none of these cases was any other cause for marked enlargement of the spleen found. From this series of cases, then, it would seem that, although a palpable spleen is fairly uncommon in pernicious anemia, it does occur sometimes, and marked enlargement may be expected occasionally.

The *liver* was enlarged in thirty-seven of our cases, exactly one out of three. It was not very pronounced in most cases, but in a few it was quite large (Table V).

TABLE V

Cm. below costal margin	No. of cases	Per cent of total
1	5	4.5
2	11	9.9
3	10	9.0
4	2	1.8
5	6	5.4
6	3	2.7

In every case a careful search for any factor to cause a chronic anemia was made. For this reason the *rectum* of each patient was examined carefully and the findings were just about as would be expected in any other series. Twenty-one patients (18.9%) had a few external hemorrhoids, two had internal hemorrhoids which had caused them some trouble, and in the rest the rectum was normal. One patient had undergone a hemorrhoidectomy.

Opthalmoscopic examinations were made on seventy-seven patients. It is recorded as absolutely normal in eight. In fifty-three minor abnormalties of no interest in connection with pernicious anemia were mentioned. The remaining sixteen showed hemorrhages, in one eye alone in six and in both eyes in ten. Rather severe hemorrhages were noted in two of these latter. Hemorrhages in the retina were encountered in those patients who from other features were evidently in the later stages of the disease.

Oedema was present at one time or another in thirty-one (27.9%) of these patients. In all of these it was found on admission and in all, except those very seriously ill, it cleared up during the first few days in the hospital.

Examination of the nervous system revealed no abnormality in twenty-nine patients; but in the remaining eighty-two (73.9%) disturbances either in sensation or in the reflexes were encountered. Sixty patients (54.1%) had disturbances in sensation, and seventy-eight (70.2%) had sluggish or absent reflexes. Nine (8.1%) had a positive Babinski sign. Several patients with subjective nervous symptoms had no abnormality of reflexes, but only three had objective signs without subjective symptoms. The disturbances of sensation recorded for each one of these patients was loss of vibratory sense when tested over the feet. Thirteen patients had some abnormality of reflexes without any demonstrable objective disturbance of sensation. In the majority of these patients, however, the variation of the reflexes from normal was slight.

Blood

It is difficult to study in tabular form the blood picture found in a series of patients with pernicious anemia, and quite impossible to present in percentages all the different features encountered. The degrees of poikilocytosis, anisocytosis, polychromasia, etc., may not be given absolute values, for they are not capable of quantitative determination, and only general statements may be made. Quantitative studies of the number of red blood cells, the quantity of hemoglobin, the number of platelets and different forms of white blood cells can be carried out, and more definite values for them may be presented.

Examination of the red blood cell counts and hemoglobin readings indicate that, once pernicious anemia has manifested itself sufficiently to bring the patients to a physician, the anemia persists. Among those of our patients who had just begun to feel badly and had reported to the hospital very early, there were many who responded excellently to treatment with extensive and prompt remissions, and some who, when discharged from the hospital, looked like normal persons and felt well. Yet in all the red blood cell counts done on these 111 patients not one of 5 millions was recorded and only two of 4½ millions. These last counts were obtained on patients just before discharge from the hospital, one a man and one a woman. There were eleven patients who, when discharged, had red cell counts between 4 and 4½ millions, making only a total of thirteen (11.7%) who, when discharged after treatment, did not have red blood cell counts lower than 4 millions. The red blood cell counts on admission show that pernicious anemia is a disease almost always associated with a very severe anemia. Thus sixty-six (60.3%) of our patients had red blood cell counts under 2 millions, and ninety-six (87.3%) under 3 millions. There is certainly no other condition commonly associated with anemia in which such severe grades of anemia are so constant.

As is to be expected in a chronic anemia of slow development, the reduction in *hemoglobin* does not show an equivalent grade of severity. Among our patients there were ten (9%) upon admission, and thirty-six (32.8%) upon discharge, who had hemoglobin values over 70%. Tabulated in detail the red blood cell counts and hemoglobin determinations appear as follows (Table VI).

TABLE VI

FIRST R. B. C. COUNT			LAST R. B. C. COUNT	
Count	No. of cases	Per cent of total	No. of cases	Per cent of total
½ m.—1 m.	12	10.8	2	1.8
1 m.—1 ½ m.	32	29.7	9	8.1
1 ½ m.—2 m.	22	19.8	15	13.5
2 m.—2 ½ m.	18	16.2	12	10.9
2 ½ m.—3 m.	12	10.8	19	17.1
3 m.—3 ½ m.	8	7.2	24	21.6
3 ½ m.—4 m.	2	1.8	13	11.7
4 m.—4 ½ m.	4	3.6	11	9.9
4 ½ m.—5 m.			2	1.8
FIRST HEMOGLOBIN READING			LAST HEMOGLOBIN READING	
Per cent	No. of cases	Per cent of total	No. of cases	Per cent of total
10— 20	12	10.8	1	.9
20— 30	18	16.2	4	3.6
30— 40	27	24.6	9	8.1
40— 50	17	15.3	19	17.1
50— 60	13	11.7	19	17.1
60— 70	14	12.6	19	17.1
70— 80	4	3.6	20	18.0
80— 90	6	5.4	10	9.0
90—100	0	0	4	3.6
100— —	0	0	2	1.8

Before discussing the *color index* it is necessary to say a word about the colorimeters used. All hemoglobin determinations were made on Sahli instruments, each one of which had been standardized in the clinical laboratory by one of the authors, so that a patient with 14 grams of hemoglobin per 100 c.c. of blood would read 90%. The normal hemoglobin readings on these instruments, therefore, was between 80 and 90%, and one might expect as normal color indices from 0.8 to 0.9, 0.9 being the upper limit of normal. With these instruments there were on admission but eight (7.2%) of our patients with color indices lower than 0.9, or the upper limit of normal, leaving one hundred and three (92.8%) showing color indices higher than normal. On discharge eleven patients (10%) showed color indices that were not above normal. Among all the patients, there were only two on admission and none on discharge who showed color indices that could be considered low normals, and both of these patients had so little hemoglobin that it could not be accurately measured. As these patients improved and got enough hemoglobin to record accurately, their color indices were above 1.0. Several patients showed very high color indices; one with 1.7, another with 1.6, and four with 1.5. Pernicious anemia shows, therefore, a high color index as a constantly characteristic feature, as is to be expected in any condition with an anemia of such long duration and slow development.

Anisocytosis and *poikilocytosis* were present in every blood film examined. The grade of anisocytosis and poikilocytosis varied more or less independently of each other, but each was more marked when the anemia was severe than when it was of a milder degree. In the more pronounced grades of anemia, usually both, but always one of these features was marked. If only one was very marked, it was poikilocytosis in a distinct majority. The study of these cases leads one to the conclusion that in the presence of a severe anemia, if poikilocytosis and anisocytosis are not frankly present, the condition surely is not pernicious anemia; and if poikilocytosis is not easily recognizable some other diagnosis must be sought. With the mildest anemia there is always an appreciable amount of anisocytosis and usually some poikilocytosis.

Basophilia is rarer than poikilocytosis or anisocytosis. Many patients with all grades of anemia show no basophilia at all. Punctate basophilia is very rare. Diffuse basophilia is most common during periods of regeneration, but even then it is by no means constant. When it does occur, it is seen in all types of cells, but possibly more frequently in large regular cells than in small, distorted poikilocytes. Contrary to the popular opinion, the findings in these cases would indicate that basophilia is not common enough in pernicious anemia to be a helpful diagnostic feature. Indeed, it is the authors' opinion that basophilia is more often encountered in certain secondary anemias than in pernicious anemia.

Nucleated red blood cells were seen at least in one film during the period of study and treatment in sixty-five cases (58.5%). They were not seen in any one of several blood films in forty-six cases (41.4%). Usually as the minimum in each case, four or five films on different days were examined. One case showed no nucleated red blood cells in seventeen preparations on different days, one in thirteen, and two in twelve preparations. Among the sixty-five cases in which nucleated red blood cells were present, there were ten in which they were seen in each blood film examined. The frequency with which nucleated red blood cells were found appeared to have no relation to the stage of the disease or the severity of the condition at the time the blood films were made. Thus, one patient admitted with 1,344,000 red blood cells and 28% of hemoglobin and discharged with 2,088,000 and 40% of hemoglobin showed nucleated red blood cells in each one of the five blood films examined; whereas another patient admitted with 1,184,000 red blood cells and 21% of hemoglobin, and discharged with 2,488,000 red blood cells and 46% of hemoglobin, showed no nucleated forms in five films examined. Among the less severe cases, one patient admitted with 3,230,000 red blood cells and 49% of hemoglobin, and discharged with 4,144,000 red blood cells and 60% of hemoglobin, had nucleated red blood cells in six of nine films examined; while another admitted with 3,290,000 red blood cells and 65% of hemoglobin, and dis-

charged with 3,370,000 red blood cells and 75% of hemoglobin, had no nucleated red cells in eight films examined. From the data afforded by these case histories, however, it is very apparent that nucleated red blood cells are found most frequently and are most numerous in patients showing the greatest improvement. Hence, although they do not help one in deciding upon the stage of the disease, they do aid to some extent in making a prognosis for the immediate future. This applies, however, only when the nucleated red blood cells seen are predominantly normoblastic. If megaloblastic, or if among the nucleated red blood cells seen there are no normoblasts, the finding is of contrary significance.

One of the most outstanding features of the blood picture in pernicious anemia is a *leucopenia*. The average of the highest white blood cell counts recorded at any time on each of the 111 patients in this series is 7,363. This is the more remarkable because it includes counts on some patients at a time when they had a transient leucocytosis associated with infections, as follows (Table VII):

TABLE VII.

Leucocyte count	No. of cases
Between 20 and 21,000	1
Between 20 and 19,000	1
Between 17 and 18,000	1
Between 14 and 15,000	3
Between 13 and 14,000	1
Between 12 and 13,000	5
Between 11 and 12,000	3

The average of the lowest white blood cell counts recorded on all patients is 3,058. This in itself is striking enough, but on studying those patients who at one time or another had a higher count, we found that practically all of them had a leucopenia at one time in the hospital. There were only eleven patients out of the entire group who did not have at one time or another less than 5,000 white blood cells; there were but six who always had more than 6,000; three never fell below 7,000, but none maintained a white blood cell count of 8,000 or above during their study in the hospital. Very low counts may be expected; and indeed, a patient with anemia and even a mild leucocytosis, in the absence of an infection, probably does not have pernicious anemia. If there is a leucocytosis associated with a very severe anemia, he certainly does not.

The *differential* white blood cell formula in pernicious anemia is interesting. The polymorphonuclear neutrophile cells are diminished, the average percentage in all the counts on all the patients being 55.8. The average number of polymorphonuclear neutrophile cells found in the differential counts made was below 65% in eighty-three (74.8%) of our patients. It was below 60% in sixty-eight (61.3%). In only ten patients was the average of the polymorphonuclear neutrophile cells over 70%, and, in eight of these, counts are included which were made during the leucocytosis of severe infections. A good percentage of polymorphonuclear cells can occur in pernicious anemia, for there was one differential count showing a polymorphonuclear neutrophile percentage of over 70 in fifty-six patients (50%). Accordingly, although a polymorphonuclear neutrophile leucopenia is a characteristic feature of pernicious anemia, an increased percentage of polymorphonuclear neutrophile cells in one count does not preclude the possibility of this diagnosis. On the other hand, sustained polymorphonuclear neutrophile leucocytosis, especially in the absence of a demonstrable infection, renders it quite unlikely.

Eosinophilia is not a characteristic feature of pernicious anemia. The average number of eosinophiles found in all the differential white blood cell counts done on all our patients was 1.58%. There were only seven patients (6.3%) who had an average eosinophile count of over 5%, and only twenty (18%) who showed an average eosinophile count of over 3%. Among these latter, however, there were several very high counts recorded; one each of 17%, 16.7%, 15%, 14%, 12%, 10.5%, and two each of 13%, 10%, 9.2% and 9%. These single counts, of course, bring up the average and make even more striking the low figures given above. These statistics compare very well with the findings of Levine and Ladd [1] and show that, although at times really numerous eosinophiles may be present in pernicious anemia, they are for the most part not increased, so that eosinophilia is not a characteristic feature of the disease. The presence of an eosinophilia was, in our cases, not related to any other feature of the disease, and was, therefore, of no prognostic import.

The number of *large mononuclears* (not lymphocytes) and transitional cells was essentially normal. The average of those found in all the counts on all our cases was 5.2%. There were four patients in whom the average of these cells was quite high—between 14 and 16%—but only five others, or nine in all (8.1%), with more than 9%. Apparently, therefore, one may say that, although high counts of mononuclears and transitionals may occur, they are not characteristic.

Immature white blood cells may be encountered occasionally in the peripheral blood of patients with pernicious anemia. Myelocytes were seen at one time or another in thirty-nine (35.1%) of our patients. They were seen more than once in 22, four or more times in 9 cases. Usually there were only a few, but in several cases 5% were recorded, in a few 6%, in one 8%, and in another 10%. Immature lymphoid forms, pathological lymphocytes, were seen in twelve cases (10%). The highest number of these cells recorded is 6%, and in but one of the twelve cases were they observed in more than one blood film.

Platelet counts were not made in these cases, but a note on each blood film was made as to whether they were absent, markedly diminished, slightly diminished, normal, or increased. In no blood film examined were the platelets noted as increased. A few were described as showing a normal number, but in most of these it is qualified by the term "low" normal. In all but these very few cases the platelets were noted as diminished, and when the anemia was severe, as markedly diminished or absent. Even in the presence of a leucocytosis the platelets did not seem to be increased. A diminished number of platelets is therefore a characteristic feature of this disease.

Wassermann Test.—The diagnosis of pernicious anemia in the presence of a positive Wassermann test is at best somewhat uncertain. Two of our patients, however, gave a positive reaction. One of these gave a single plus twice and a negative on the third test; and in the other a suggestive positive on the blood was recorded twice and a negative test on the spinal fluid was obtained.

The statistics gathered for this report are of no value in determining the frequency with which syphilis and pernicious anemia are found associated. But one might here emphasize the fact that a diagnosis of pernicious anemia is never justified in the presence of a positive Wassermann test until the findings of pernicious anemia have been shown to exist after a thorough course of antiluetic treatment.

Urine Analysis.—Studies of our cases reveal the fact that pathological findings in the urine in pernicious anemia are very common. Sixty-one (54.9%) of our patients showed albumin in the urine, most of these on more than one occasion. It was not excessive in any case, but in all but four there was an appreciable amount. In four, just recognizable traces were recorded. Casts, however, were a rare finding. In all the urine analyses done on ninety (81%) of the patients, none were found. In the remaining twenty-one, casts were found, hyaline only in eleven and both hyaline and granular in ten cases. In none were the casts found in great numbers. Urobilin was found in the urine of fifty-six (50%) of our patients. In almost all of these cases in which it was recorded once, it was found repeatedly. Only a few cases showing urobilin at all failed to show it throughout their stay in the hospital. As is to be expected, urobilin was most frequently found in those patients who were in bad condition and who did not react promptly and extensively to hospital treatment. Other urinary findings were of little interest. Bile was present in three analyses, blood in two, acetone in six. Sugar and diacetic acid were never encountered.

Gastric Analysis.—The findings on analysis of the gastric contents of our patients were very interesting. *Not one analysis on any of these patients showed any free hydrochloric acid.* Complete achylia gastrica was not a common finding, but a low total acidity was the rule. The combined acid was below 10 acidity per cent in ninety-four cases (84.7%). It was below 20 acidity per cent in all but four cases. These cases showed combined acidities of 33, 30, 28, and 25. Lactic acid was recorded in the gastric contents in three cases. Blood was a more common finding. The microscopic examinations showed nothing of interest.

According to our analyses, then, the absence of free hydrochloric acid in the stomach contents is as constant a finding in pernicious anemia as is the anemia itself. These conclusions are also supported by the report of Levine and Ladd,[1] who found in 107 patients with pernicious anemia only three with free hydrochloric acid in the stomach contents. One of these patients presented a most uncertain diagnosis of pernicious anemia, and another had three analyses of stomach contents with no free hydrochloric acid, the fourth with only 6%. If these exceptions are kept in mind and the cases of Levine and Ladd are added to ours, we have 218 cases of undoubted pernicious anemia, with only one showing an appreciable amount of free hydrochloric acid in the gastric contents.

Stool Examination.—The stools of all but thirteen patients were examined and showed nothing of special diagnostic interest. Blood was found present, as determined by the guaiac test, in the stools of nine patients on one examination. For each one of these patients a stool examination is listed in which negative guaiac tests were obtained. Bile was marked positive in the stool examinations in twenty-one patients (21.4%). Cysts of *Amoeba coli* were recorded once, and flagellates noted three times. The patient showing *Amoeba coli* cysts was one of the three in whose stool flagellates were encountered.

Temperature.—No elevations of temperature above normal were recorded in fifty-three cases (47.8%) during the stay in the hospital. In addition there were twenty-one patients who had no fever except following transfusion. These added to the former make seventy-four patients (66.6%), who had no fever due to the pernicious anemia itself. Four other patients had no fever until just before death, when it became very high. The remaining thirty-three patients (29.7%) had some fever with no demonstrable cause other than the pernicious anemia. The fever in these patients varied irregularly between normal and 100° F., occasionally going as high as 101° F., but rarely above this point. These latter patients with slight fever from time to time did not, as a rule, feel as well as those without it, but this was the only difference. Those who during the first week in the hospital had some fever seemed to progress and improve just as well as those who were afebrile.

SUMMARY

In the above report, the case histories of 111 patients with pernicious anemia are analyzed. It is impossible to present in summary the results of this study, but a

few of the points demonstrated by it, which probably need emphasis, may be mentioned.

1. Pernicious anemia is about equally common in men and women, or possibly, slightly more common in men.

2. It is a disease of adult life. It is never seen in children, is uncommon in old age, and occurs most frequently between the ages of 40 and 60 years.

3. It is very uncommon in negroes.

4. The anemia, once established, practically never completely regenerates.

5. Anisocytosis and poikilocytosis are constant findings. Basophilia is rather uncommon, although it does occur.

6. Although a leucocytosis may occur in the presence of an infection, a leucopenia is the characteristic finding.

7. An eosinophilia as high as 17% may be found, but an actual increase in eosinophile cells are the rare exception. The same applies to the large mononuclear and transition cells.

8. Myelocytes, as high as 10%, may be found in an occasional case.

9. Free hydrochloric acid in the gastric contents of these patients is so rare that, when present, it should put the diagnosis under suspicion.

10. The spleen is palpable in only about 20% of pernicious anemia patients. It may, however, extend 5 to 6 cm. below the costal margin.

11. The liver is palpable in about 33% of pernicious anemia patients. The edge may be palpated 5 to 6 cm. below the costal margin.

12. All patients, whatever other discomforts they may have, suffer from one or more of the following symptoms: weakness, disturbances of digestion, and nervous disorders of the extremities.

The authors wish to thank Dr. Winford H. Smith and Dr. Warfield T. Longcape for permission to use the material on which this report is based; and Miss Fannie Riach for her courteous cooperation in the history room of the hospital.

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A CHEMICAL STUDY OF A CASE OF CHLOROFORM POISONING

By H. J. Stander

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In 1919, Davis and Whipple carefully reviewed the question of delayed chloroform poisoning. The experiments of Whipple and his coworkers show that a protective action against liver injury from chloroform anaesthesia can be exerted by certain food-stuffs as well as by some drugs and chemical agents. Such a protection can be induced by feeding the animals, in the days preceding the anaesthesia, sugar and diets rich in carbohydrates, extracts of liver or kidney, beef extract, brain, skim milk as well as commercial casein. The authors conclude that no single theory will explain this protective action of these food substances and that it must be a reaction of the liver cells rather than of substances circulating in the blood stream. (This final conclusion does not seem quite justified in the face of the experiments performed.) They also found a marked susceptibility to liver injury in starved animals and in animals that had received fat, or combinations of food containing fat in large proportion. According to them, then, the view that the "fixing" of chloroform by fat results in a greater injury to the cells in certain localities will not serve to explain the increased susceptibility noted in starved animals. In further support of this argument it may be noted that brain, a substance quite rich in lipoids, appears in their list of foodstuffs that exerted a protective action.

These workers further demonstrated that glucose, or cream, given intravenously during chloroform anaesthesia, does not modify the effect of the drug on starved animals. A similar result was noted in the case of hypertonic solutions of sodium carbonate as well as phosphate solutions given intravenously during the anaesthesia. Before discussing these results, I shall report a case of delayed chloroform poisoning that recently occurred in our clinic.

Case Report

C. S.—Colored, housewife, aged 18. Admitted October 1, 1923.

Family and Personal History.—Negative. The general health had always been good.

Present History.—The patient was admitted to the hospital at about the end of the ninth month of pregnancy. Her menstrual history had been normal. Measurements taken in the Obstetrical Dispensary at about the seventh month of gestation revealed a generally contracted rhachitic funnel pelvis; the measurements being: Spines 22, Crests 24, Trochanters 27.5, Baudelocque's Diameter 17, Diagonal Conjugate 11, and Tubera Ischii 7.5 cm. The pubic arch was narrow. The sacrum had a

definite false promontory and its vertical concavity was markedly increased.

Physical Examination.—In brief, a healthy young woman. Wassermann negative.

Examination of the abdomen revealed a nine months' pregnancy with the child in R. O. P., head floating, and foetal heart in R. L. Q. Measurements of the pelvic outlet were: Anterior Sagittal 5.5; Posterior Sagittal 11, and Anteroposterior Diameter 12.5 cm.

History at Time of Labor.—On October 22, 1923, at 2.45 P. M., the patient was admitted to the delivery-room with a history of having had pains since 6 P. M. the previous day. Rectal examination showed the head one cm. above spines, membranes intact, canal not obliterated, and cervix 2 cm. dilated. Pains fair but irregular. At 7.35 P. M. condition unchanged except cervix 3 cm. dilated. Several rectal examinations from this time till 9.30 P. M., October 23rd, showed very slight progress. The head had descended to 1 cm. below spines, but had progressed no further during the last three hours, with the cervix staying about 6 cm. dilated. At 9.45 P. M. the membranes were ruptured artificially. The head appeared at the outlet at 9.50 P. M. At 10.45 P. M., October 22nd, as the head had made no further progress, and in view of the funnel pelvis, it was decided to terminate the labor artificially.

Operative History.—On vaginal examination it was found that a thick caput was responsible for the fact that the scalp was visible at the outlet. The head had just reached the level of the spines. With the whole hand in the vagina the position was found to be R. O. T. After manual dilatation of the outlet, the blades of the forceps were applied to the sides of the head. After rotation of the occiput anteriorly the head was brought to the outlet by intermittent traction. When the vulva was well distended the blades were removed and delivery completed by Ritgen's Manoeuver.

There was a slight bilateral cervical tear and a moderate second degree laceration of the perineum, both of which were repaired with catgut. The placenta was expressed from the vagina, presenting by the Schultze Method. Bleeding was estimated at 200 c.c.

The baby breathed spontaneously at birth. The head showed a large caput and was slightly moulded.

Anaesthetic.—Chloroform. Started 11.06 P. M.; ended 12.15 A. M. Operation begun 11.15 P. M.; child delivered 11.33 P. M.; placenta delivered 11.57 P. M.; operation ended 12.20 A. M. Amount of chloroform used 2 ounces.

Pituitrin 1 c.c., after completion of third stage.

Course of Puerperium.—The day following delivery, the patient's condition was normal, but on October 24, 1923, her temperature was about 100° F. all day, while the pulse ranged between 130 and 140, and was weak but regular. Respirations varied between 24 to 32. She seemed rather drowsy and slow in answering questions, and vomited late that evening.

October 25, 1923.—The temperature rose steadily from 100° F. at 8 A. M. to 102.5° F. at 4 P. M.; the pulse increased from 130 to 150, and the respirations from 24 to 52. At 4 P. M. a physical examination showed the patient totally uncooperative, and in a much agitated condition. She complained of pain under the right chest. The mucous membranes were cyanosed and the sclerae jaundiced. She became comatose during the examination. Heart sounds clear, pulse 150 per minute. Lungs clear except for a small area about 6 cm. in diameter, in the right interscapular region between the 4th and 8th vertebrae, where the breath-sounds were somewhat distant, a few fine râles were heard, and the percussion note was higher in pitch than over the surrounding lung tissue.

The upper limit of liver dullness was in the 4th interspace; and the edge of the liver was palpable 2 f. b. below the costal margin. It was very tender. Abdomen somewhat swollen and tympanitic.

8 P. M.—Infusion of 1,000 c.c. of salt solution started. The pulse became stronger.

10 P. M.—Respiration 60, pulse 160, temperature 102° F.

10.30 P. M.—A medical consultation revealed nothing in the chest.

11.50 P. M.—Respirations ceased. Adrenalin was injected into the heart through the 5th interspace, but without success.

Unfortunately no autopsy could be obtained.

Blood Count.—At 5.15 P. M., October 25, 1923, the differential count was:

W. B. C., 23,500; P. M. N., 84.5%; P. M. E., 0; P. M. B., 0; L. L., 0; S. L., 7.5%; L. M. & T., 3%.

ROUTINE EXAMINATION OF URINE

Date	Character	Sediment	Sp. Gr.	Reaction	Albumin	Sugar	Microscopic
10/ 4/23	Clear	+	1.009	Neutral	v.s.t.	0	Negative
10/11/23	Cloudy	+	1.012	Acid	s.t.	0	Epithelial cells +++ W.B.C. ++
10/20/23	Cloudy	+	1.010	Neutral	v.s.t.	0	Many epithelial cells, and W. B. C.

As soon as the patient presented the signs and symptoms of a toxemia, the following analyses were made:

URINE

I.—A single catheterized specimen at 8 A. M., Oct. 25, 1923.
Volume, 200 c.c.
Total Nitrogen 1.56 gms., or 7.8 gms. per liter.
Urea Nitrogen 1.07 gms., or 68.5% of Total Nitrogen.
Ammonia Nitrogen 0.08 gms., or 5.1% of Total Nitrogen.

II.—A single catheterized specimen at 4 P. M., Oct. 25, 1923.
Volume, 235 c.c.
Total Nitrogen 1.69 gms., or 7.2 gms. per liter.
Urea Nitrogen 0.49 gms. or 28.9% of Total Nitrogen.
Ammonia Nitrogen 0.15 gms. or 8.8% of Total Nitrogen.

BLOOD

Taken at 4 P. M., October 25, 1923.
Non-Protein Nitrogen 65.2 mgms. per 100 c.c. of blood.
Urea Nitrogen ..... 37.36 mgms. per 100 c.c. of blood.
Uric Acid ..... 12.2 mgms. per 100 c.c. of blood.
Chlorides .....536.0 mgms. per 100 c.c. of blood.
Sugar ..... 58.0 mgms. per 100 c.c. of blood.
Amino Acids ..... 12.02 mgms. per 100 c.c. of blood.
CO_2 Combining Power—27.4 vols. per cent.

Since 1894 there have been three other cases of delayed chloroform poisoning in this clinic. Two of these were fatal and showed typical central necrosis of the liver. Comparing the histories of the four cases, we find:—

1. All the patients underwent an operative delivery lasting one hour or more, from two and one-half to four ounces of chloroform being administered during this period.

2. The first symptom of poisoning in all cases is nausea followed by vomiting, which appears from 18 to 24 hours after the administration of the anaesthetic.

3. Shortly after the vomiting starts, jaundice appears and soon becomes marked.

4. The patient then becomes either drowsy and later comatose, or first irrational, quite violent, and then sinks into a comatose state.

5. The temperature, respiration or pulse are no index of the severity of the condition except, perhaps, near the end. The heart seems to function fairly normally.

6. An enlarged, tender liver is present in all cases.

7. There is a leukocytosis of about 20,000 in all cases.

8. In the three fatal cases, death ensued from 2 to 6 days after the administration of the drug.

9. Our treatment in the non-fatal case differed from that in the fatal cases in that the patient was given calcium lactate, grs. xv q. 2 h., and later 5% glucose solution by the Murphy method. She received approximately 2,500 c.c. of this solution.

10. It should also be noted that in the non-fatal case, the kidneys continued to function far better than in the other cases as evidenced by the urinary output.

Discussion

The urine and blood analyses, as given above, furnish us with the most interesting and perhaps most important information on this fatal case. These findings may be summed up as follows:

(1). An increased protein metabolism, as shown by the increased output of nitrogen in the urine.

(2). A very marked decrease of the urea-nitrogen percentage in the urine, due undoubtedly to an impairment of the urea-forming function of the liver, and perhaps partially to a raising of the urea-threshold in the kidney as shown by the rise in the blood urea-nitrogen.

(3). A steadily rising ammonia coefficient in the urine. This, of course, is to be expected, as the rate of conversion of ammonia into urea in the liver is greatly retarded. It is also possible that the greater portion of the ammonia in the urine serves as a base to transfer acid radicles formed within the body.

(4). A very definite drop in the CO_2 combining power of the blood, indicating an acidosis. The formation of HCl from chloroform, suggested by Graham as a cause of the acidosis, does not seem quite probable. To determine whether this is a true acidosis, it is necessary to know the CO_2 tension, the pH or the concentration of the H_2CO_3. Van Slyke has shown that the values of the concentration of $BHCO_3$, in conjunction with a factor such as the pH, yield information concerning the available alkali, which might be quite difficult to ascertain in any other way.

(5). The increase in the non-protein nitrogen of the blood, as well as the marked rise in its uric acid content, would indicate a retention resulting from renal insufficiency. It may be well, however, to bear in mind that an increased disintegration process of the purine bodies will result in a greater production of uric acid.

(6). The liberation of amino acids from the breakdown of body protein is accelerated and the conversion in the liver of amino acids into urea retarded, resulting in an accumulation of amino acids in the blood stream.

(7). A marked fall in the blood sugar content. This is probably associated with the liver injury.

The above changes indicate an increased destruction but incomplete oxidation of the proteins—a disturbance of the liver function together with a moderate degree of renal insufficiency.

It has been shown that the blood corpuscles contain more chloroform than the blood plasma during, or immediately after, the administration of this drug. It also seems true that the richer a cell is in fat content the more chloroform it will contain, owing no doubt to the solubility coefficient of chloroform for lipoidal substances. The chloroform must to a certain extent be "fixed" by all or some of the lipoids, and therefore will not be readily excreted. This would explain the marked susceptibility to liver injury from chloroform following a diet rich in fats. In the case reported above, the patient was on a regular ward diet during the days preceding the administration of chloroform, and at that time received approximately 170 grams of fat, 370 grams of carbohydrate and 130 grams of protein.

From the results following brain-feeding, as reported by Davis and Whipple, it would seem essential to know just which lipoids are most important in this "fixing," or whether there is another substance in brain which offsets this process. I cannot agree with the authors in holding that their results following starvation disprove Opie's theory of chloroform "fixing" by fats, for in starvation the proteins are already being destroyed, normal metabolism no longer exists, and the factor of low fat content is not the only one to be considered.

Summary

A study of the cases of delayed chloroform poisoning that have occurred in this clinic, taken in conjunction with the experimental work of Opie, Whipple, and others, strongly indicates that a diet rich in carbohydrates and milk, and restricted in fats, is advisable before chloroform anaesthesia is resorted to. That the use of alkalies is of benefit is still doubtful. To obtain any further helpful information on the mechanism of chloroform poisoning, it seems necessary to study the distribution of the various lipoid-substances during this condition. The effect of chloroform anaesthesia on the blood-sugar level appears to be of significance. The association between hypoglycemia and liver injury is still obscure and certainly worthy of further experimental work.

I wish to express my thanks to Dr. W. A. Perlzweig for his assistance in one of the analyses.

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DIFFUSE UTERINE GLAND CYSTS IN A CAT

By A. F. GUTTMACHER

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In this note I wish to describe a very interesting and unusual pathological specimen found in an autopsy on an old female cat. The lesions were entirely localized to the reproductive tract. Here were found: a large hydrometra, diffuse glandular cysts of the endometrium, bilateral cystic ovaries, and a complete obliteration of the cervical canal. Such an extensive formation of endometrial cysts is probably unknown for the human. The added complication of a completely obliterated cervical canal and the resulting hydrometra makes the specimen even more uncommon. In Kelly and Cullen [1] a myoma of the uterus is pictured with four or five small uterine cysts, which on histological examination appear to be simple retention cysts of the uterine glands. Anspach [2] mentions the uncommon condition of glandular hypertrophy with cystic degeneration which occurs after chronic endometritis, most commonly in the senile. W. L. Williams [3] pictures and describes a case, similar in part to ours, in the cow. Lockyer [4] speaks of endometritis polyposa cystica and considers it to be of inflammatory origin. Cullen [5] describes a case of pyometra in the cat; in this instance there were no endometrial cysts. The writer has not attempted to undertake a comprehensive review of the literature.

Clinical history.—Female cat, age 14 to 15 years.

Past history, negative for diseases. Numerous pregnancies, the last occurring about five years before the animal was autopsied. No known complications of the pregnancies.

Present illness.—About three years ago there was noticed, by the family who owned the cat, a symmetrical swelling of the abdomen, which exactly resembled a normally progressing pregnancy. The abdomen continued to swell for about three months when the cat suddenly disappeared, to return twenty-four hours later with an abdomen of normal size. It was thought by the family that the cat had secretly borne and deserted her litter. This unprecedented happening occurred twice again, during the next two years.* About six months before the animal was autopsied, the abdomen began to enlarge for the fourth time. This enlargement was permanent and became increasingly greater. The cat was suffering markedly and was brought to this laboratory to be killed.

AUTOPSY

The uterus looked as large as one at term; both cornua were equally distended. The uterine vessels were large and conspicuous, the whole picture simulating a pregnancy. The abdominal cavity was otherwise normal. The serosa of the uterus was smooth and glistening. There was no free fluid in the abdomen. On opening the uterus, about one and a half litres of clear, odorless, brownish fluid were recovered. There were no particles of fibrin, no gross evidence of pus in the fluid.

The rest of the abdominal and the thoracic contents were macroscopically negative. There were no abnormalities of the urinary tract.

Gross specimen.—Uterus, right cornu—12 cm. in length, 10 cm. in circumference. The endometrium is splotchily discolored with areas of recent and old subepithelial haemorrhage. Almost the entire endometrium is replaced by diffuse cysts—single and coalescent. On first glance, because of the vesicles, the endometrium reminds one of the skin in a severe case of dermatitis venenata. The lesion is so widespread that there is not an area of 2 sq.

* It is altogether probable that the cat gradually developed an increasing hydrometra which finally burst its way through the cervical canal.

mm. that is not covered by a cyst. The single cysts range from fifty micra to 10 mm. in diameter. The coalescing cysts are quite large; the largest just above the internal os is 22 by 13 by 11 mm. On section many present the typical honeycomb arrangement of the familiar multilocular cysts of the ovary. Some cysts contain colorless fluid; while in others the fluid is brown-tinted.

Left cornu—13 cm. in length, circumference 11 cm. The endometrium is the same as in the right horn except that there is no evidence of haemorrhage. All the cysts of the left horn contain only clear colorless fluid.

Internal os.—The endometrial cysts are continuous directly into the cone-shaped internal os, several medium-sized cysts occupying the apex of the cone. There is no evidence of a cervical canal.

Cervix.—On transverse section through the middle of the cervix, no canal is seen nor is any found on searching with a probe. Progressively concentric narrowing rings of white dense tissue completely occlude the lumen.

External os.—The external os is represented by a shallow dimple which is continuous with the occluded canal.

Vagina.—Normal. No cysts, no haemorrhage.

Tubes.—Both tubes are apparently normal. No cysts of the epithelium.

Ovaries.—The left ovary is of normal size and quite solid. There are numerous small macroscopic subserous cysts, none more than one millimeter in diameter. No large follicles nor corpora lutea are seen.

The right ovary is twice as large as the left. It contains only a thin framework of solid tissue, in which are embedded eight clear fluid containing cysts, varying from 2 to 9 mm. in diameter.

Microscopic

Uterus.—The right cornu. The normal architecture of the uterine endometrium is largely altered by diffuse simple, glandular cysts in all stages of formation. In the space between these variously-sized cysts there remain many completely normal uterine glands. All transition stages from these normal glands to large tense cysts are seen. It appears as though the process was in full activity at the time the animal was killed and new cysts and larger cysts were continually being formed. The epithelium lining the smaller cysts is of the usual columnar ciliated and non-ciliated varieties. In the larger cysts, the greater intracystic pressure has flattened the epithelial cells to a thin single low layer. Many of the individual epithelial cells of the normal glands and smaller cysts have the histologic appearance that we commonly associate with secretory activity. The fluid contents of the cysts do not stain; under the microscope the cavities appear like empty spaces. There is no histological evidence of endometrial hyperplasia or of a neoplasm. There is some thickening of the subepithelial connective tissue, which is difficult to see because of the compression of the stroma by the cysts. The subepithelial stroma is the site of moderately recent and of very fresh haemorrhage; the haemorrhage extends down to the myometrium. The myometrium is normal except for fatty infiltration into the deeper layers. No myoma is present. The multilocular coalescing endometrial cysts can be differentiated from similar cysts of the ovary only by the remaining intercystic stroma. Some of the larger cysts have normal glands and smaller cysts budding off from their wall. The haemorrhage into the subepithelial stroma is also present in the walls of the coalescing cysts—here the haemorrhage is relatively quite old.

The lesions of the left cornu are similar to those of the right, except that there is no subepithelial haemorrhage.

Cervix.—The cervical canal has disappeared—there is no vestige of epithelium to mark its former site. The center of the narrowing periluminal concentric muscle bundles is occupied by a dense area of old vascularized scar tissue about 350 by 250 micra. By means of the Van Gieson stain it is demonstrable that this complete obliteration of the cervical canal has been brought about through the double mechanism of smooth muscle hypertrophy at the periphery of the above area and of dense fibrous scarring in the center. There are no signs of an active inflammatory condition. Stains for bacteria are uniformly negative.

Tubes.—The extreme periphery of the tubal lumen contains little follicles, or loculi, formed by an adherence of the plicae to each other.* There is also a slight increase in the thickness and density of the periluminal connective tissue. The epithelium is normal and ciliated.

Ovaries.—Both ovaries have numerous simple, follicular cysts. There are no maturing Graafian follicles, nor are there any corpora lutea. The interstitial cells are well developed, in their morphology resembling human paralutein cells. The tissue is not like that of typically senile ovaries, because there is no fibrous scarring, and because Graafian follicles in various stages of development are present.

Discussion

This condition of diffuse endometrial cyst formation must be very rare even in the cat. For on specific interrogation of the other members of this department and of many workers in allied departments no one has observed

* In the human this is interpreted as an aftermath of a previous inflammatory process.

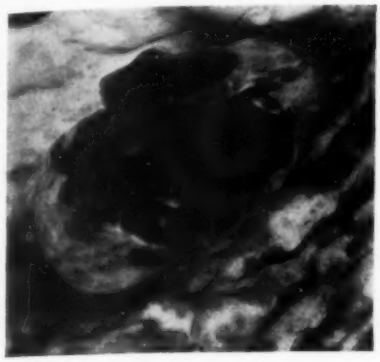

Fig. 1

Fig. 2

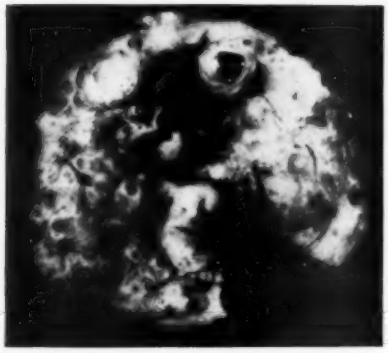

Fig. 3

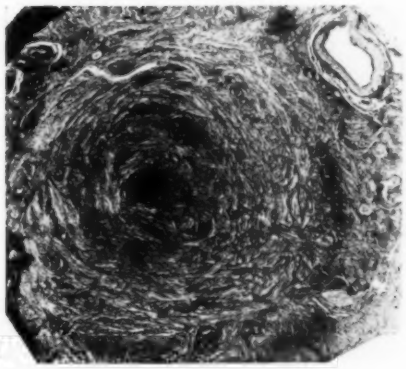

Fig. 4

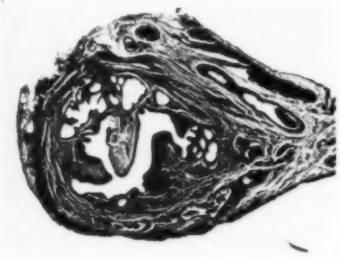

Fig. 5

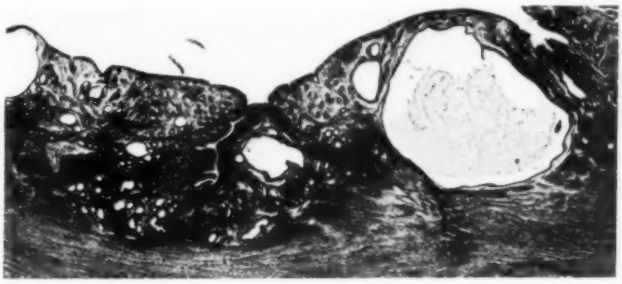

Fig. 6

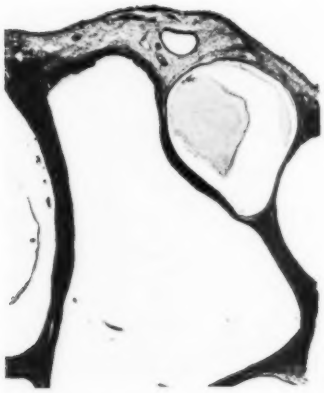

Fig. 7

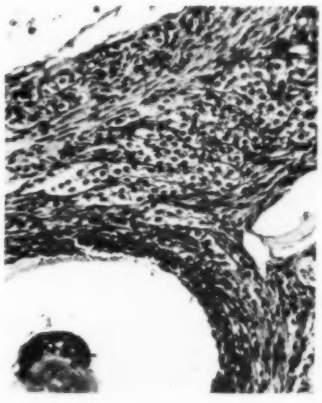

Fig. 8

or heard of a similar case.* In a personal communication Dr. Frederick L. Reichert has informed me that a few years ago he had a senile cat in which there was a huge hydrometra, the abdomen touching the ground; but no observations were made regarding the presence of cysts.

For the aetiology of this pathological process one can only offer negative and unsatisfactory explanations. There are certain points, however, which are incontestable: the cysts of the uterus are simple glandular retention cysts; through some means the mouths of the uterine glands have been completely occluded and the gland secretion retained. Also we may be certain that the cervix has been totally obliterated by a process of scar formation. The hydrometra is probably either uterine gland secretion which has undergone some change and has been retained *in utero* because of the occluded cervix; or it is a simple intrauterine transudate of the cyst fluid through the walls of the cysts.

There are two possible explanations that occur to us for the sealing over of the gland ostia and the occlusion of the cervix. The one possibility is senile fibrosis, and the other post-inflammatory scarring.

Let us critically examine the possibility that senile fibrosis is the cause of the pathological picture with which we are dealing. Marshall [6] states that cats have been known to bear young in extreme cases at the age of 14 years; but he makes no statement of the average age at which reproduction ceases. Our animal at the time this process was first noted was about twelve years old, hence the possibility of senile changes and its attendant fibrous scarring and atrophy cannot be disregarded. However, to contradict this theory is the fact that the ovaries show neither senile fibrosis nor atresia. Since the cat has but two or three sexual seasons a year the absence of maturing follicles is of no value in dating our animal in the reproductive life-cycle. The uterus shows no hypoplasia, and only very mild subepithelial fibrosis. There is a mild grade of arterio-sclerosis in the uterine and cervical arteries. We therefore have no conclusive evidence that the reproductive tract of this cat had begun to show senile changes.

We are convinced from our study of the material that there was no active inflammatory condition at the time the animal was killed. All we see of this puzzling process is the result: retention gland cysts, obliterated cervix and small tubal loculi. There is no mononuclear invasion; however, there is moderate thickening of the subepithelial connective tissue in both uterine cornua and tubes. It is conceivable that we are dealing with a well healed post-inflammatory process which has left no more evidence than this.

I should like to thank Dr. G. B. Wislocki for permitting me to study the specimen, and also for the encouragement and guidance he has given me during this study. I should also like to express my appreciation to Dr. J. W. Williams and Dr. W. G. MacCallum, Dr. T. S. Cullen and Dr. E. Novak who discussed the specimen with me.

EXPLANATION OF PLATE

Figs. 1, 2, 3, are gross photographs × 2.

Figs 4-8, are photomicrographs H. & E.

Fig. 1.—Portion of uterine endometrium from right cornu showing diffuse gland cysts. In center large multilocular

* The following extraordinarily interesting case report which appeared after this note had gone to press was brought to my attention through the kindness of Dr. J. W. Williams. The similarity of the pathological condition in this patient with the lesion in the cat's uterus is obvious.

"Ein fall von exzessiver cystischer Hyperplasia des gesamten Endometrium." U. Westphal. Zentralblatt fur Gynäkologie, 1923, Nummer 44, S. 1707. Hamburg. Dr. Westphal's patient first presented herself at the clinic, Feb., 1922. She was 39, married, and had never been pregnant. She had had a leucorrhoeal discharge for one year. Following her previous menstrual period, 3 weeks before admission, a bloody vaginal discharge was noticed, for the relief of which she sought medical aid.

Her general physical examination was negative. Pelvic examination: From the external os there presented a rough cauliflower-like, mushroom-shaped polyp the size of a mandarin. It was of soft consistency and dark blue red in color. The pedicle of the tumor was soft, elastic and narrow; it was continuous with the internal os. The uterus was not enlarged; adnexa, normal.

During the examination traction was made on the polyp which tore its pedicle; there was no bleeding from the severed pedicle. On bimanual palpation it was felt that the pedicle of the tumor originated from the fundus. On microscopic study the whole tumor showed marked cystic dilatation of low cylindrically-lined epithelial glands, the glands being separated by scanty non-characteristic stroma. The patient was discharged and not heard from until one year later when she returned with the same complaint of slight bleeding.

Pelvic examination. The uterus was anteflexed, of usual size and consistency. The cervix was markedly dilated. The examining finger was able to palpate the surface of the whole endometrium which felt diffusely nodular. Some tissue was excised for study; it showed a morphology similar to that of the polyp excised the year before. On the basis of these findings it was thought wise to remove the uterus.

Gross specimen. The myometrium is slightly thickened; there is a sharp line of demarcation between the myometrium and the much hypertrophied endometrium which measures from one to three centimeters. The whole endometrium is replaced by cysts varying from a pinpoint to a walnut in size.

Microscopic. Myometrium normal. The cysts are lined with cylindrical epithelium which arises from a true basal membrane. Several of these cystic dilated glands contain a rose-colored homogenous substance. The interglandular stroma is much compressed, varying from a hair to a needle in thickness. There is a slight amount of round cell infiltration. This transformation of the endometrium ends sharply at the internal os.

In his brief discussion Dr. Westphal suggests, but rules out, the possibility of cystadenoma. The presence of the leucorrhoea and the long history of sterility made Dr. Westphal favor a chronic endometritis, as the cause of the cyst formation, although there is no conclusive histologic picture to support this view. He claims that his case of diffuse endometrial cysts is the first ever to have been reported.

cyst which has been cut open to show loculi. Note areas of subepithelial haemorrhage.

Fig. 2.—Area of uterine endometrium from right cornu.

Fig. 3.—Endometrium in region of internal os, showing uterine gland cysts descending into apex of cone. A triangular block of tissue removed for microscopic section from periphery.

Fig. 4.—Cross-section of cervix. × 10. Cervical canal obliterated by scar tissue. No vestige of lumen remains.

Fig. 5.—Right tube in region of isthmus. × 14. Lumen shown with abnormal tubal follicle or loculus.

Fig. 6.—Uterus. Right cornu. × 13. Section shows diffuseness of process also the variation in size of cysts.

Fig. 7.—Multilocular endometrial gland cyst of right cornu. × 14.

Fig. 8.—Section of right ovary. × 140. Note interstitial cells, also resting follicle—ovum detached through artefact.

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MARKED STRUCTURAL ALTERATION IN A BREAST CARCINOMA RECURRING AFTER ELEVEN YEARS

By Edward M. Hanrahan, Jr.

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The following case presents a striking example of certain unusual manifestations of neoplasia, namely, malignant transformation of the stroma or very marked alteration of epithelial cells, in a long delayed recurrence of a breast carcinoma.

The patient, an unmarried white woman, forty-nine years old, was admitted to the Union Protestant Infirmary April 13, 1910, complaining of a "sore breast." There was a history of trauma six years before admission, followed by the appearance of a small lump in the upper and inner quadrant. The lump persisted, although the patient was unaware of any increase in its size. Two years later, about four years before coming to the hospital, she consulted a physician, who, however, did not suggest surgical treatment at that time. Six months before admission she began to have pain, the breast and skin became sore about the nipple and a sanguino-serous discharge appeared. Ointments were used without effect. On examination at the time of operation the left nipple was seen to be retracted and the adjacent skin indurated and covered with crusts. The skin, except just over the infected area, was freely movable. There was neither pigskin retraction nor dimpling. The axillary glands were enlarged and tender. A radical operation—removal of the breast, with both pectoral muscles and a clearing out of the axilla—was performed by Dr. J. M. T. Finney.

The post-operative course was complicated by a phlebitis of the left leg.

The tumor was examined at the time by Dr. J. C. Bloodgood and by others, who agreed in a diagnosis of Paget's Disease of the breast.

About 11 years later the patient noticed a small lump which appeared over the ribs just medial to her operation scar. It was hard and adherent to the ribs, but gave rise to no pain. The skin itself was apparently normal. This lump grew slowly for two months. Three weeks before the second admission to the hospital the patient was operated upon in a small country hospital. She stated that her doctor had thought the lump to be an abscess, and she was afterwards told that it was not completely removed. The wound healed, but the lump soon reappeared and grew rapidly. On examination there was seen, in addition to the old operative incision, a small scar behind the left shoulder and one beneath the right breast due to the removal of two small tumors twenty years previously.

Just medial to the line of incision for removal of the left breast, over the third and fourth ribs, there was a purplish red tumor mass about the size of a lemon, standing well out from the surrounding chest wall. The skin was tense, shiny and seemed ready to ulcerate. It was surmounted by a well healed scar of a recent incision. There was fluctuation over the summit of the mass, but the base was firm and hard. There were no other nodules. There were no masses palpable in the axilla or beneath the clavicle.

The heart was not appreciably enlarged, although the mediastinal dullness seemed to be slightly increased. The liver edge was just palpable. No x-ray examination of the chest was made at this time.

The patient was operated upon by Dr. H. L. Homer. A cautery was used for the excision, but the growth was found to extend down beneath the ribs and was connected with the pleura. Only a partial excision was done, with the expectation that radium would be used immediately.

After six days, at the time when the patient took her first radium treatment at the Howard A. Kelly Hospital,

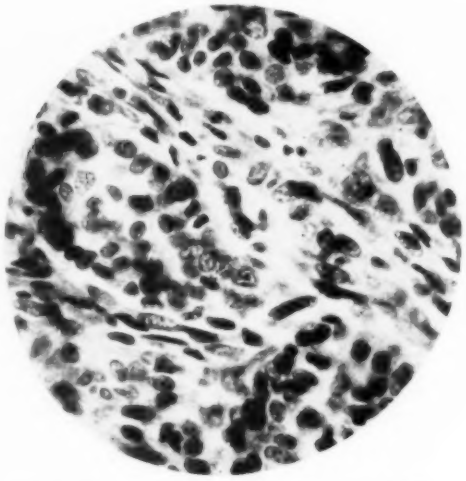

Fig. 1.—Recurrence of breast carcinoma after 11 years. Section shows definite epithelial island in the midst of spindle cells.

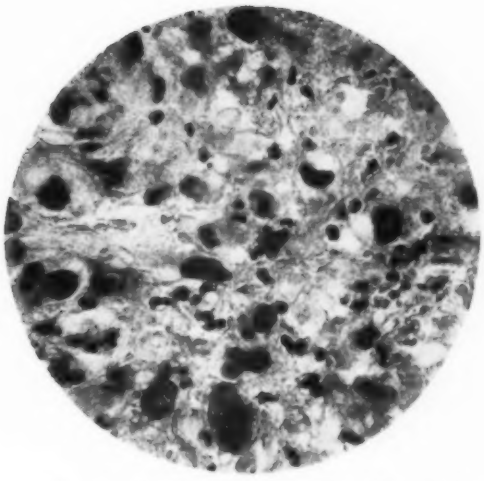

Fig. 2.—Recurrence shown in Fig. 1 three weeks after radium treatment. Section shows large irregular epithelioid cells.

Fig. 3.—Popliteal tumor of second case showing its mixed character.

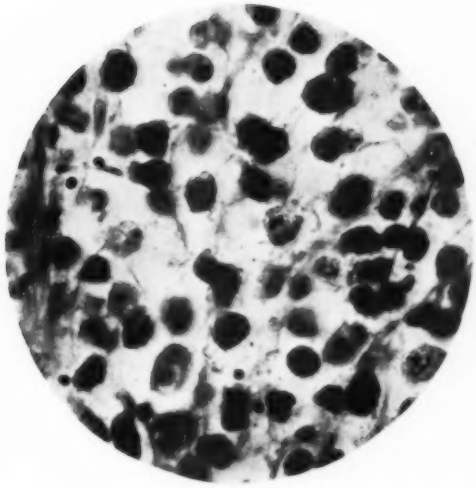

Fig. 4.—Femoral tumor made up of large round cells.

it was evident that the tissue in the open wound had grown rapidly. During the following three weeks the growth appeared first to be checked and later regressing. An x-ray made at this time showed a mass in the mediastinum projecting out into the left lung, the appearance of which suggested a new growth. The patient was submitted to another radium treatment after three weeks, with some subsequent improvement in the appearance of the wound. About five weeks after the operation a cough developed, followed after another week by dyspnoea. From this time on her general condition became gradually worse and the tumor and granulation tissue grew rapidly. The patient died four months after operation. No autopsy was performed.

This case presents two interesting features. First, the late development of a recurrence; and second, the morphology of the growth. Here, only the second will be considered.

At operation the growth presented the typical appearance of "fish flesh." It was soft and custard-like. The diagnosis from frozen sections made at this time was sarcoma. Paraffine sections, which were more satisfactory, showed the tumor to be very cellular and made up of large and small irregularly-shaped spindle cells. The nuclei were also variable in shape and size, but for the most part small and elongated. They were arranged irregularly. These cells and their arrangement had the appearance of a sarcoma. There were, however, in addition, islands and areas of perfectly typical epithelioid cells, which were arranged in the sheet-like fashion of squamous-cell carcinoma with scant intercellular substance. Both cells and nuclei were large. Many cells contained two or more nucleoli and mitoses were numerous (Fig. 1).

Sections taken three weeks after the first radium treatment showed quite a different picture. The spindle cells first described had entirely disappeared, and most of the section was made up of necrotic eosin-staining material in which were many polymorphonuclear neutrophiles and lymphocytes. There were also scattered, large, swollen cells resembling the epithelioid cells just described. They stained more intensely with hæmatoxylin and showed fewer mitotic figures but more nucleoli and vacuoles. There were many "bird's-eye inclusions" (Fig. 2).

Dr. W. G. MacCallum, after examining these sections, called attention to their resemblance to certain tumors described by Ehrlich and Apolant, which they had observed in their studies on propagable tumors, and which showed a hitherto undescribed property of tumor growth. After ten transplantations of a mouse adenocarcinoma, the stroma began to manifest properties of unrestrained extension and to show a spindle-cell growth suggesting sarcoma.

As is well known, occasional recurrences and metastases are met with which show a marked divergence from the original growth. Metastases may show greater or less restraint than the parent growth; or they may present a microscopical appearance suggesting that in the case of a mixed tumor only one element has metastasized or at least has retained in the metastasis the power of independent growth.

This characteristic was well illustrated in another of our cases. The patient presented herself with two tumors, one popliteal, the other femoral. Both tumors—popliteal and femoral—were removed at the same operation. Microscopic examination of the popliteal tumor showed it to be a mixed-cell sarcoma made up of large round and spindle cells. The femoral tumor had an entirely different appearance, being made up exclusively of large round cells. The femoral tumor was considered to be a metastasis from the popliteal, but made up of only one element of the parent growth. The only other interpretation seemed to us to be that the popliteal tumor was made up of a single type of cell which was capable of occurring in two quite different forms. We felt, however, that such marked variation in morphology was less to be expected than the metastasis of but one element of a mixed cell tumor.

Recurrences also show alteration in structure and, as Ewing says, more especially after operation. But whether this transformation of epithelial cells is ever sufficient to produce a picture identical with spindle-cell sarcoma is questionable. Loeb, Woglom, Lewin, Bashford and Russel recorded the development of sarcoma in propagable carcinoma of rats and mice.

Ewing cites the three theories which have been offered in explanation of the apparent appearance of sarcoma in mouse carcinoma. First, that the growths were originally mixed. Second, that the sarcoma represents a neoplastic transformation of the stroma of the host caused by a stimulation of stroma cells of the epithelium. The third —and the one favored by him—is that the spindle cells are altered epithelium. After examining several of Woglom's cases he concluded that the spindle cells were derivatives of epithelium.

Dr. Ewing was kind enough to examine the sections of our first case. He emphasized the fact that the tumor was originally a Paget's carcinoma, a disease of squamous cell epithelium, which is notoriously prone to produce spindle cells and to resemble sarcoma. He felt that the recurrence was undoubtedly epithelial and alveolar, and raised the question, "Why make an assumption in order to prove another assumption, viz., that the tumor is mixed and the stroma cells have become sarcomatous?" He preferred the simpler explanation that the spindle cells are altered epithelium, and that after radium have less capacity for alteration.

On the other hand, it may be suggested that we have not in this case an alteration or transformation of the whole tumor. There are definite spindle cells and equally

definite epithelioid cells. Is it safe to assume that some of the epithelioid cells are transformed into spindle cells and that some are not?

It is known that the effect of radium varies within the various types of new growths and more so between one type and another. While Ewing assumes that the appearance in this case after radium is due to the effect of this treatment in lessening the capacity of the epithelioid cells for alteration, the possibility must be considered that it may be an expression of the difference in the two types of cells, the spindle cells being much more affected by the radium than the epithelioid cells.

It appears desirable to report this case in view of the possibility that subsequent work may prove the fact that the stroma of a carcinoma may take on malignant properties and the resulting picture be that of a true carcino-sarcoma. Ewing admits the occurrence of primary cellular carcino-sarcomas and believes that they come from the "Anlage" which usually goes to make up an adeno-fibroma.

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ADRENAL INSUFFICIENCY AND ATROPHY OF THE CORTEX FOLLOWING VENOUS OBSTRUCTION OF THE SUPRARENAL GLANDS

By Robert Elman and Philip Rothman

(From the Department of Surgery, Hunterian Laboratory, Johns Hopkins University)

The controversy which followed Brown-Séquard's classical observation that prompt and characteristic death followed experimental removal of both adrenal glands, resulted in countless experiments. These have, in general, abundantly confirmed Brown-Séquard's original discovery. The recognition that various forms of degenerative changes occur in the adrenals of patients suffering from Addison's disease was followed by numerous attempts to reproduce this condition experimentally. All such efforts have been unsuccessful. The actual removal of only part of the adrenal tissue permits, up to a certain point, complete recovery.[1] Removal beyond this limit is followed by immediate death, quite as if all the tissue were excised. The intermediate stage which would result in death as a consequence of a very gradual and chronic insufficiency seems unlikely of production by surgical ablation.

Crowe and Wislocki[2] attempted to bring about this type of disease by the implantation of tiny tubes of radium emanation in the substance of the glands of dogs and cats. They succeeded, it is true, in producing death from adrenal insufficiency three to four weeks after the operation. The autopsies, however, showed tissue changes in neighboring structures, quite apart from the alteration in the glands themselves. With the same end in view, Marine and his co-workers[3] have used an ethyl chloride spray to freeze the cortex of the adrenals, producing insufficiency followed by death in several weeks. The animals chosen for these studies were rabbits, in which the frequent irregular occurrence of accessory cortical tissue makes it rather difficult to estimate the effects of any procedure performed on the two glands alone. Russo-Giliberti and Di Mattei[4] were unsuccessful in their attempts to produce experimental Addison's disease by cauterization, crushing, and injection of substances into the gland. Using quite another means, Hartman and Blatz[5] have recently reported that death occurs in periods of one to four months following venous ligation of the suprarenal glands in cats. They attributed the deaths of their animals to a chronic adrenal insufficiency. However, one's criteria for such a death must be quite definite, and from the description furnished and the microscopic findings reported, one remains unconvinced that the deaths were not due to other causes entirely.

The method of venous ligation has been used in the past rather freely. Indeed, many experimenters considered, as equivalent to double adrenalectomy, the ligation of the two lumbo-adrenal venous trunks. The validity

of such assumption has been discussed by Cannon,[6] who concludes that simple venous ligation may quite readily lead to erroneous results. Martinotti [7] studied the effects of venous occlusion of both central lumbo-adrenal veins. He found vacuolated and shrunken cortical cells twenty-four hours after occlusion, with no changes in the medulla. In animals which survived two to three months, the medulla and most of the cortex were replaced by connective tissue. His experiments were unfortunately carried out on rabbits and the results are somewhat confusing. It seemed of interest, therefore, to attempt the production of chronic adrenal insufficiency by venous obstruction.

METHODS

The choice of animals in experiments on adrenal glands is quite important, for the species difference is often marked. In the past this has been overlooked, and, consequently, confusing and fundamentally different results have been reported. For our own purposes cats, in general, appeared to be most suitable. They rarely possess accessory cortical bodies, so that alterations made or observed in the two glands may be safely taken as representing the whole of the cortical adrenal tissue. Moreover, the lumbo-adrenal vein is easily exposed and ligated, while the isolated position of the glands, and their technical ease of approach, are also factors in their favor. Primary ether anaesthesia was used throughout, and the usual aseptic technic observed. A right or left rectus incision was chosen, in order to avoid the midline preperitoneal fat, and to obtain firmer closure of the wound in several layers. The single incision, furthermore, made easy the exposure and manipulation of both glands. The entire operation averaged one-half to three-quarters of an hour. The behavior of the animals was thereafter observed as frequently as possible, and the pulse rate, temperature, respiration, and weight recorded.

In view of the reported production of adrenal insufficiency by simply ligating the large lumbo-adrenal vein, several animals were subjected to this procedure. In one series, bilateral ligation was performed and, in another, unilateral ligation with the removal of the other gland. The vein arises in the lumbar muscles, crosses the adrenal gland on its ventral surface with which it becomes adherent, and from which it receives several venous channels, and empties directly into the vena cava, or, as frequently happens on the left, into the renal vein. The ligation, therefore, of this large vessel at the outer and inner edge of the gland certainly should obstruct most, if not all, of the venous return. It was early noted, however, that such a ligation never caused any immediate change in the appearance of the gland. No swelling or congestion was ever produced, and it was apparent that many small veins were acting as collaterals, and that complete venous obstruction was not being caused. These channels, according to Cow,[8] leave the gland at the lower, and to a lesser extent at the upper pole. They form a direct vascular connection between the adrenal and the kidney capsule.

In another series of animals, therefore, these small veins at the upper and lower pole were cut across by blunt dissection. Slight bleeding stopped immediately; it was not necessary to employ ligatures. The tying-off of the lumbo-adrenal vein now brought about an entirely different picture. Immediate swelling and congestion developed, and from its engorgement with blood the normally pink gland changed to a dark blue. In effect, such a procedure merely obstructed the venous channels, leaving the arterial and nerve supply intact, since these latter enter the gland through its dorsal surface. During the course of these operations attempts were made to cause a partial obstruction by leaving some smaller veins intact. It soon became apparent that such an intermediate stage could not be created. Either the gland became congested and swollen, or it retained its normal appearance. The outcome of the operation, as will later be pointed out, depended entirely on which of these conditions resulted.

EXPERIMENTAL FINDINGS AND DISCUSSION

Simple ligation of the lumbo-adrenal vein.—Contrary to the assumption of a number of experimenters, ligation of the large lumbo-adrenal vein caused no demonstrable effect in the animal. Even ligations of one vein, with removal of the other gland, invariably resulted in recovery. And when the animal was killed two, three, or four months later, no apparent change could be seen at the autopsy. There was, to be sure, a moderate hypertrophy of the remaining gland, and the veins at the upper and lower poles were dilated. Its gross and microscopic structure differed in no way from normal.

Complete venous obstruction of one gland, with removal of the other.—The production of swelling and congestion in the gland following the severing of the vessels at both poles, with ligation of the lumbo-adrenal vein, was taken to indicate a complete venous obstruction. This operation was performed in a number of cats with removal of the other gland. These animals all developed signs of adrenal insufficiency and died in from three to six days.

The clinical course of such an animal is quite characteristic. The recovery from operation is usually prompt, and three to four hours later the animal is apparently normal. Very soon, however, apathy develops and evidences of asthenia and listlessness come on. Nausea, anorexia and gastro-intestinal irritability are early noted. In a few cases there was a definite irregularity in the rate and force of the pulse. A marked degree of muscular prostration always preceded death. No pigmentation was ever observed. The duration of these symptoms may be short, beginning twenty-four hours before exitus. Often, however, it is more gradual, beginning the day

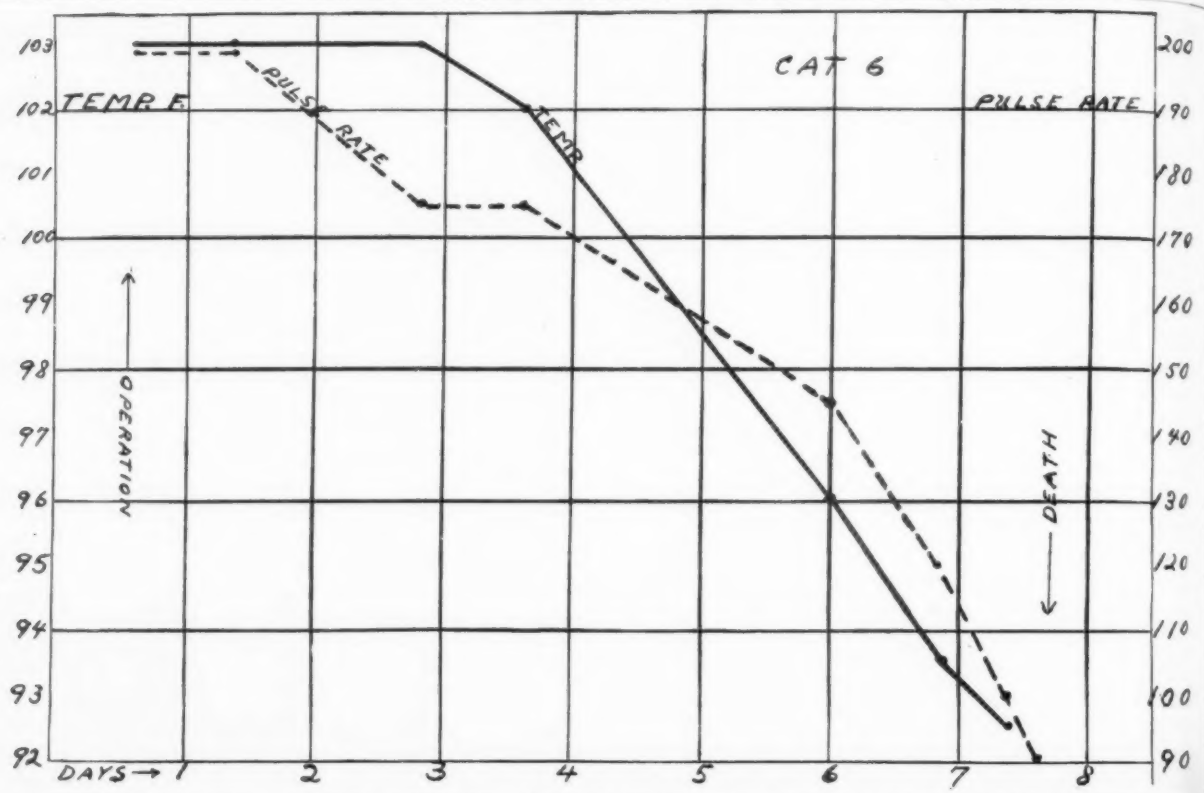

Chart 1.—Temperature and Pulse Rate Curves, following removal of one gland and complete venous obstruction of the other, namely, ligation of the lumbo-adrenal vein and destruction of the smaller veins of the upper and lower pole by blunt dissection. The pulse rate and temperature fell synchronously.

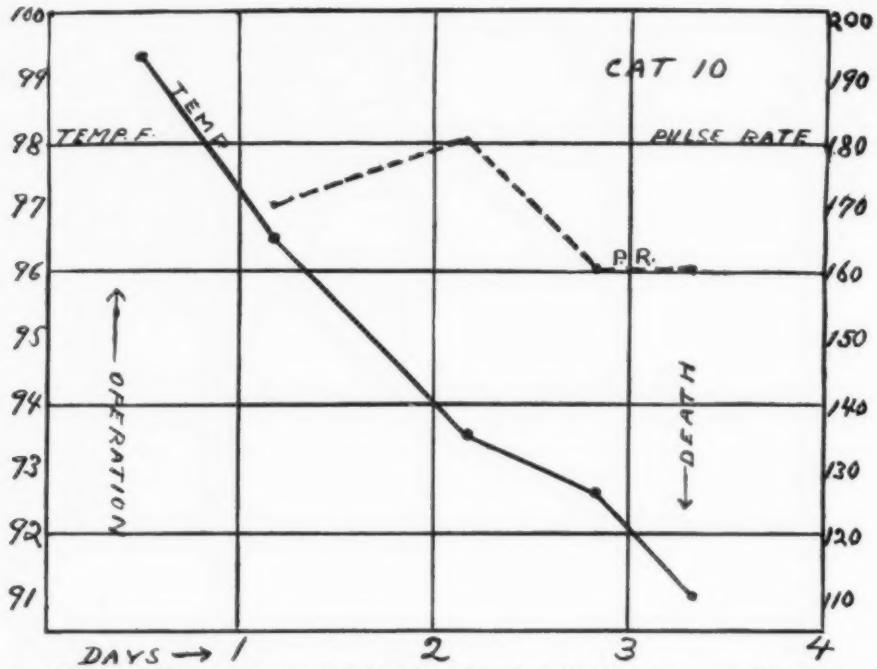

Chart 2.—Temperature and Pulse Rate Curves, following same operation as described in Chart 1. Although the pulse rate is represented as not having a terminal fall, it became impalpable before death.

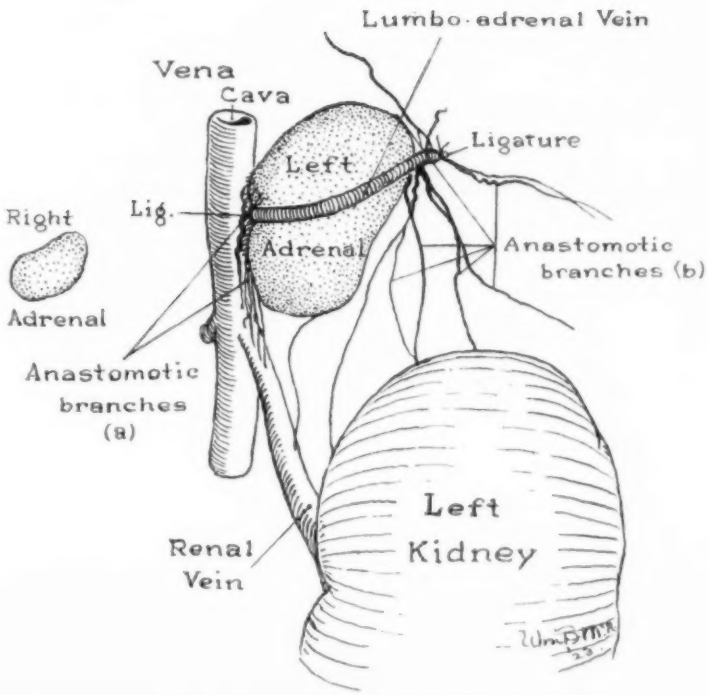

Fig. 1.—Complete venous obstruction of both glands (see text). No evidence of adrenal insufficiency were observed. The animal was killed three months later, injected arterially with India ink, and a block containing the adrenals cleared by the Spalteholz method. The drawing is made from a photograph of the specimen, and shows a marked hypertrophy on one side in contrast to an atrophy on the other. (The kidney is represented as smaller than its actual size.) Anastomotic branches are shown inside the medial ligature (A) and lateral ligature (B).

Fig. 2.—Complete venous obstruction of both glands was carried out as described. Three months later an exploratory laparotomy was done. One gland was found to be hypertrophied, the other seemed quite small. The former was removed, and death followed in twenty-eight hours. This section is from the remaining atrophied gland, and shows the only part of the tissue which contains any cortical cells (C). The medulla (M) occupies practically the entire gland. The necrotic cortical tissue is indicated (N).

after operation and increasing from two to five days till death.

In all cases the temperature and pulse rate were measured; of the two the former was found to act more constantly. Its fall always meant a lethal outcome. The pulse rate often fell with the temperature, as shown by Chart 1, but not invariably; an example of this latter condition is shown in Chart 2. The pulse always became weak with the development of asthenia, and for a period of hours before death was often impalpable, even over the precordium.

At autopsy the suprarenal gland was swollen, dark in color, and on section showed numerous hemorrhagic areas. The microscopic sections revealed invariably an almost complete necrosis of the cortex, the nuclei all being pyknotic, fragmented, or karyolytic. Only occasionally were seen small stretches of normal appearing cells confined entirely to the zona glomerulosa, and occupying the very outer edge of the cortex just under the capsule. In contrast to this extensive necrosis of the cortex, the medulla often presented areas of normal appearing tissue, with definite chromaffine staining cells. In no case had the process of nuclear destruction proceeded as far in the medulla as in the cortex. Moreover, in several specimens injected arterially with India ink, the existence of vessels in the medulla could always be seen, while the rich capillary plexus of the cortex remained uninjected. In the glands of animals dying after six days, several areas of regeneration tissue were seen surrounding the more vascularized zones of the medulla. These regions were indicated by the presence of vessels filled with India ink injection. The distribution of fat in these necrotic glands, as shown by stain with Sudan III, was always characteristic. It was confined sharply to the peripheral third of the zona fasciculata. The glomerular zone was uninvolved, as was likewise the central two-thirds of the cortex. This contrasted markedly with the normal distribution of fat through the entire broad zona fasciculata, with occasional droplets in the zona glomerulosa.

There seems to be no adequate explanation for this postponement of adrenal death from three to six days, for in our experience total removal of both glands in cats is followed by a fatal outcome within twenty-four hours. This method seemed promising enough for the production of a more chronic state by perhaps obstructing a little less completely the venous channels. As indicated before, however, the line was too sharply drawn, and the outcome was always a complete survival in cases where no congestion appeared at operation, or a typical death in six days —in cases where venous stasis and swelling were produced. The outcome could, thus, always be predicted at the time of operation, and no intermediate stage, as far as our observations went, seemed possible. Quite apart from its theoretical interest, this one-stage operation has proved useful for the study of other features of adrenal insufficiency where it is necessary to make observations over a period of days. Previously, a two-stage operation, removal of the two glands at intervals, was the only method of producing even a short chronic insufficiency.

Complete venous obstruction of both glands.—In four animals congestion and swelling in both glands at the time of operation—the arbitrary criteria used to indicate complete venous obstruction in these experiments—were produced by dissection of both poles and ligation of the large lumbo-adrenal veins. The outcome in all of these cases was an indefinite survival; no evidences of adrenal insufficiency were observed. Two of these animals were killed after several months, and an unexpected condition was found at autopsy. In both cases, one of the adrenal glands was hypertrophied and seemed twice its normal size. Injection with India ink showed the presence of collateral venous channels emptying into the renal vein and kidney cortex, as shown in Fig. 1. Microscopically, normal structure and arrangement of medulla and cortex were made out.

The remaining gland, curiously enough, was quite small, and on section showed abundant brown chromaffine-staining tissue. The light yellowish cortex seemed absent. Microscopically, it was seen that this atrophied gland consisted nearly entirely of medullary cells, only a few areas of cortical tissue being present. Thus, quite by chance, a method seemed available for producing atrophy confined to the cortex. It was apparently an accidental matter on which side the hypertrophy took place, for in one case it was on the left and in the other on the right.

The remaining two animals were subsequently operated upon, the difference in the size of the two glands similarly observed, and only the hypertrophied one removed. Both animals died, one in twenty-eight hours, the other in eight days. The undisturbed atrophied gland from the first animal was carefully studied, and a section is reproduced from the only part of the gland which showed any cortical tissue at all (Fig. 2). Fig. 2 clearly shows the striking contrast between the normally appearing medulla and the necrotic cortex.

The vital function of the adrenal gland, for a long time, has been ascribed to the production of adrenalin by its medulla. Recently evidence has been accumulating to show that this assumption is untenable. Biedl[9] has demonstrated that in elasmobranch fish in which the cortical substance exists as a separate structure, the interrenal bodies, extirpation of this tissue is followed by death. Crowe and Wislocki,[1] who carefully studied the effects of partial or complete removal of the adrenals in mammals, record their opinion that it is the cortex that is essential to life. Wheeler and Vincent[10] entirely removed the left adrenal, amputated one-half of the right, and cauterized the remaining portion of the medulla.

Many of their animals survived. Houssay and Lewis,[11] working with dogs, removed the medullary portion of both adrenals by opening the gland lengthwise and scooping it out with a spoon. The glands were sewed up with catgut, and all the animals lived. If the remaining cortex was removed later on, death occurred. From our experiments described above, additional evidence is presented concerning the essentialness of the cortex. In the two cases in which death followed destruction of the cortex, the remaining medulla stained deeply with chromic acid, thus indicating that it had retained its normal reaction, although in itself, unable to maintain life.

Summary

1. No symptoms or signs of adrenal insufficiency result from simple ligation of the lumbo-adrenal vein, even with removal of one gland. Indefinite survival follows such a procedure.

2. A method is presented for the production of adrenal insufficiency, with death in three to six days, following a one-stage operation in which one gland is removed and the veins of the other obstructed.

3. Atrophy confined to the cortex, and sparing the medulla, always occurred in one adrenal gland, after bilateral ligation of the lumbo-adrenal vein and blunt dissection of the vessels of the upper and lower poles. The other gland was hypertrophied, but otherwise normal.

4. Further evidence of the non-vital function of the medulla is presented in the production of a prompt death following the removal of all the cortical tissue, in spite of the existence of a normally-staining medulla.

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MELANURIA IN THE ABSENCE OF MELANOTIC TUMOR

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The melanins constitute a group of brown and black pigments formed in many vertebrates and invertebrates. They occur as the normal pigments of the hair, the choroid coat of the eye, and the skin of man, and are present in large amounts in the ink-sac of certain cephalopods, notably the squid. The black pigments in many worms and insects are melanins. Likewise, the blackening of certain fungi on exposure to air is due to the development of melanins.

Melanin is not a chemical entity of definite and constant composition. As a group, the pigments are characterized by their high carbon and low nitrogen content and the variable, though usually high, sulphur content. They are probably very complex compounds. Gortner[1] has suggested that the apparent variation in the chemical composition of melanins may be largely due to the method of isolation employed. The strong alkalis which are used to dissolve the pigments probably often cause alterations in their chemical structure. Analyses of the isolated pigments, then, are apt to give varying results.

The pigments are amorphous, insoluble in water, weak acids, alcohol, ether, and other ordinary solvents. They are soluble in alkalis. The black pigments occurring normally in man result from the activity of specialized cells. The exact manner of formation, and especially the relative parts played by nucleus and cytoplasm of the cell, are still a matter of dispute. Under certain abnormal conditions melanin may be formed in the entire absence of cellular activity.

The chemical changes involved in the formation of a melanin are a matter of interest. It is known that such substances are formed only from certain aromatic groups of the protein molecule, such as tyrosin and tryptophane. It seems that an oxyphenyl group is at least necessary for the nucleus of a melanin. Simple phenol compounds, homogentisic acid, tyrosin, and tyrosin-containing polypeptides may all give rise on oxidation to brown or black pigments. The parent substances may be represented by the formula: R. C_6H_4.OH, where R is H or some complicated group. Eppinger,[2] found that a patient with melanuria excreted more melanin when fed tryptophane. He considers that tryptophane usually forms the nucleus of melanin.

The actual conversion of the mother substance into melanin is effected through the agency of the ferment tyrosinase, which is widely distributed in animal and plant tissues.[3] This ferment is plentiful in the animal tissues in which melanin is formed, such as in the ink-sac of the squid and the skin of the negro. It is widely distributed in the plant kingdom. It is very abundant in the fungi in which there is a production of dark pigments on oxidation, and also in insects which develop dark pigments. It seems probable that the chemical process is the same wherever melanin is formed, namely, an oxidation of cyclic complexes from the protein molecule, through the agency of tyrosinase. Von Furth and Jerusalem [4] concluded that where melanin is formed in large amounts the tyrosinase acts in conjunction with autolytic enzymes. The latter split the chromogen groups from the protein molecule which is then oxidized to melanin. It is of interest in this connection to note that in the hydrolysis of protein, by prolonged boiling with acid, colored compounds resembling melanins are formed.[5]

That melanin may occur in the urine of patients suffering from melanotic sarcoma has been recognized for almost a century. Only about 20 per cent of such patients, however, have melanuria during the course of the disease. The melanuria does not usually develop until metastases have appeared in the liver. In this condition melanin is formed in abnormally large amounts through the normal activity of specialized cells. The melanuria represents an overflow of the pigment through the kidney.

There has been much discussion as to whether true melanin may appear in the urine in conditions other than melanotic sarcoma. Bonnet [6] reports the finding of melanin in the urine of five patients suffering from secondary carcinoma of the liver (not melanotic), and in one patient with lymphatic leukemia. Litten [7] noticed it in the urine of a case of peritonitis; Gnezda [8] in a patient with tuberculosis of the intestines; and Zoeppritz [9] in stenosing carcinoma of the colon with ileus and peritonitis. Stewart [10] reports the finding of black pigment in the urine of a patient suffering from a benign pigmented tumor of the pelvis, which was considered a parovarian cyst. The breasts of this patient after an operation on the pelvic tumor and again after childbirth discharged large quantities of an inkblack fluid. Oppenheimer and Kline,[11] in reporting a case of ochronosis, state that the black pigment, found in the cartilages and often in the urine in this disease, had been identified as melanin in 3 of the 41 cases reported up to 1922. The pigment found in the urine of the case reported by them was identified by Janney [12] as melanin. Brierhoff's patient [13] voided black urine during treatment for acute urethritis.

It seems questionable whether the color of the urine noted, at least, in all the cases cited, was in reality due to melanin. Feigl and Querner,[14] in a recent review of the subject, concluded that such cases should not be accepted. These authors think that melanuria occurs only in, and is diagnostic of, melanotic sarcoma. The difficulty arises from the fact that substances other than melanin may give colored compounds with the reagents used in testing for melanin. Urine containing melanin is usually a normal color when voided since the melanin is in the form of colorless melanogen. When the specimen is allowed to stand exposed to the air, the colorless melanogen is converted into the brown or black melanin through oxidation.

The usual tests for melanin are based on the production of a colored substance on the addition of such oxidizing reagents as bromine water, nitric acid, chromic acid and ferric chloride. The most satisfactory reagent is ferric chloride, the addition of which to urine containing melanogen produces immediately a brown or black precipitate. Such a precipitate, while characteristic of melanogen, may develop in its absence owing to the presence of other oxidizable chromogens. Helman [15] after a careful study of the identification of melanin in urine decided that true melanin should be considered as present only when (a) the addition of ferric chloride gives a brown or black precipitate, which (b) dissolves on the addition of sodium carbonate, giving a brown or black-colored solution and (c) from which a brown or black amorphous powder is precipitated on the addition of a mineral acid. These requirements suggested by Helman still hold. The three reactions in sequence are specific for melanin. The melanogen in urine may also be oxidized to melanin by passing an air current through the alkalinized specimen for several hours. From this black solution Janney [12] has shown that pure melanin may be precipitated by adding 25 per cent acetic acid. Thormahlen [16] found that urine containing melanin, to which 5 per cent sodium nitroprusside and a small amount of sodium hydroxide have been added, turns blue on the addition of acetic acid as a result of the formation of Prussian blue. This reaction is, however, not positive with all urines containing melanin and may occur in the absence of melanin. In the series reported here no specimen has been considered as definitely containing melanin unless the melanin could be precipitated out by the technic of Helman.

Practically the only two conditions in which absolutely black urine occurs are melanuria and alkaptonuria. In both the urine is colorless when voided but darkens from the surface downward on standing. Substances which may be confused with melanin and the tests by which they are differentiated may be noted:

(1) *Bile pigments:* In the presence of an intense jaundice of long standing the urine is very dark. The clinical condition and the tests for bile, however, easily differentiate such urine from that containing melanin.

(2) *Blood pigments:* Where there is bleeding high up in the urinary tract the hemoglobin may be changed to methemoglobin and make the urine almost black. Tests for blood differentiate this from other types of pigments.

The ancients considered the occurrence of black urine as of the most evil prognostic significance, probably because they recognized it only when containing blood or bile pigments.

(3) *Hematoporphyrin:* This iron-free derivative of hemoglobin when present in large amounts usually gives a Burgundy red color to the urine. The tests typical of melanin are not given by this substance. It can be positively identified only by its absorption spectrum.

(4) *Indican and its oxidation products:* These may be found in the urine as a result of excessive putrefaction of protein within the intestine. When they are present in excessive amounts, the urine may be almost black from the formation of oxidation products. The urine does not blacken on the addition of ferric chloride, and tests for indican, such as Obermeyer's, are strongly positive.

(5) *Alkaptone bodies:* These are the substances responsible for the black urine of alkaptonuria. The most important is homogentisic acid. The anomaly is due to an inability of the body normally to dispose of tyrosin. Dakin [17] found, however, that in alkaptonuria there is an excessive production of homogentisic acid as well as an inability of the body to oxidize it. The urine in this condition turns black on standing. It does not blacken on the addition of ferric chloride; it stains cloth, and reduces alkaline copper solution, but not alkaline bismuth solutions.

(6) *Oxidation products of phenols:* The prolonged use of phenol may give rise to black pigments in the urine. These do not give the tests for melanin or alkaptone bodies. The diagnosis must be made largely from the history of the prolonged use of phenol.

Our interest in the subject of melanuria began with the finding of melanin in the urine of a patient suffering from intestinal obstruction. Total specimens were being preserved daily for certain chemical studies. It was noticed that they gradually turned black from the surface downwards, becoming after a few days almost jet black. Tests for bile pigments, indican, and similar bodies were negative. The freshly voided specimens, which were of a normal color, gave a black precipitate with a solution of ferric chloride, which was dissolved by an excess of sodium carbonate forming a black solution. From the solution an amorphous black powder was precipitated by hydrochloric acid. All specimens for two weeks gave positive tests for melanin, although the intensity of the reactions varied from day to day. Autopsy revealed no evidence of a tumor.

We have had the opportunity of examining the urine from four other patients with intestinal obstruction. Melanin was demonstrated in large amounts in all. One of the patients had an obstruction of the ileum, the exact cause of which was not disclosed at operation or autopsy. The obstruction in the second patient occurred subsequently to an operation for a gangrenous appendix. In another, the obstruction was due to a large fecolith at the lower end of the ileum. In the fourth patient the obstruction followed a postoperative peritonitis, due to the hemolytic streptococcus. The fifth patient was studied during one of numerous recurrent attacks of obstruction due to adhesions from a generalized peritonitis. The chemical findings in three of the cases have been made the subject of a separate report.[18]

Intestinal obstruction is an intoxication characterized chemically by a definite train of events.[19] The blood chlorides begin to fall very quickly after the obstruction occurs. After the chlorides are depleted below a certain level, a marked destruction of body protein begins. The urine then shows a very low chloride and a high nitrogen content. Usually the destruction proceeds more rapidly than the damaged kidney can excrete the excess nitrogen; hence the blood shows an increase in non-protein nitrogen. This increase is mainly in the urea nitrogen and the undetermined nitrogen. The intimate relation of the chlorides to the intoxication is emphasized by the facts that the protein destruction does not begin until the chlorides have fallen below a certain level, and that the protein destruction may be entirely prevented by providing an abundant supply of sodium chloride.[13]

There is much to suggest that the protein destruction here is due to a generalized autolysis of body tissue. It has been shown that in pyloric obstruction, a toxemia identical with that after intestinal obstruction occurs.[21] In pyloric obstruction there is a high non-protein nitrogen, low chloride and low sodium content in the blood.[22] There is every reason to believe that there is also a low sodium content in high intestinal obstruction, although this has not been experimentally determined. Bradley [23] thinks the reason, that the proteolytic ferments which are always present in the body tissues do not destroy the protein, is that the protein is normally in combination with sodium and can not be acted on by the pepsin-like ferment which must initiate the protein destruction. For the action of this ferment the sodium-protein combination must be changed into an acid salt, or into uncombined protein. When this is done, the initial cleavage of the protein may begin. Any condition which will give rise to acid sodium protein salts or uncombined protein will afford favorable conditions for the action of the proteolytic ferments, and as a result autolysis will begin.

If this view be correct, a low sodium content of the tissues would provide optimal conditions for autolysis. It seems evident also that the conditions must also be optimal for the conversion of the cyclic complexes so formed into melanin, presumably through the action of tyrosinase. It seems quite possible also that the conditions may be favorable for a production of unusually large amounts of such complexes which by oxidation will give rise to melanin. Crystals of tyrosin and leucine and some purple, needle-shaped, crystals, which could not be iden-

tified, were demonstrated in a specimen of urine from one of the patients here reported. The urine from another patient, which contained much melanin, also showed many black needle-shaped crystals which were not identified.

It has been pointed out that there are certain other diseases characterized by chemical findings similar to those found in intestinal obstruction.[19] In lobar pneumonia there is uniformly a fall in blood and urine chlorides—a phenomenon which has never been satisfactorily explained. We have examined the urine daily from two patients suffering from lobar pneumonia. One showed very large amounts of melanin during one period of the disease, the other a moderate amount. The urines of these patients were very low in chlorides and high in nitrogen.

There is much to suggest a chemical similarity of pernicious anemia to intestinal obstruction. Many facts might indicate an intestinal origin for the disease. An absence of free hydrochloric acid from the gastric juice is practically a constant finding. We have had the opportunity of studying the urine and blood of a patient suffering from pernicious anemia during a rapid relapse. The presenting symptoms were referable largely to the central nervous system, the result of a marked myelitis. The blood showed a low sodium chloride content. The chloride excretion in the urine was very low and the nitrogen excretion very high. Tests for melanin in the urine were strongly positive.

In experimental anaphylactic shock in rabbits, Major[24, 25] has shown that there is a marked rise in nitrogen excretion with an increase in the non-protein nitrogen, and a fall in the chlorides of the blood. The chloride metabolism in serum disease in a man has been studied by Rackmann, Longcope, and Peters with similar results.[26] The observation was made that specimens of urine collected from rabbits during anaphylactic shock turned black on standing. This change was found to be due to the presence of melanin in large amounts. We have examined the urine daily from rabbits injected at intervals with horse serum. Some of the specimens contained large amounts of melanin. All contained at least a trace. The urine of normal rabbits usually contains no melanin, although it seems that occasionally a small amount may be present. No opportunity to test for melanin in the urine of a patient during anaphylactic shock has presented itself.

Melanin has also been identified in the urine of a patient dying with acute pancreatitis (verified by autopsy) during the course of diabetes. The blood chlorides of this patient fell as low as 280 mgs. (normal 450-500) per 100 c.c. of blood. Similar findings were observed in another case of diabetes. The urine of this patient had a straw color when voided, but rather quickly became a grass green. This color faded and was followed by a black color which developed from above downwards. Pure melanin could be precipitated from the specimens repeatedly. The examination of the urine of numerous other patients suffering from diabetes failed to show any trace of melanin.

Haden and Orr[27] have reported in detail the chemical findings in the blood and urine of three patients during an acute reaction following gastroenterostomy. These were characteristic of intestinal obstruction. The patients probably had an obstruction at the gastroenterostomy stoma. Melanin was demonstrated in the urine of two of them. One patient at the height of the reaction showed many purple crystals in the urine, which could not be identified, as well as crystals of leucine and tyrosine.

A summary of the conditions in which melanin has been demonstrated is given in Table I.

TABLE I.

Summary of conditions in which melanuria has been noted in the absence of melanotic tumor.

DISEASE	No. cases
Intestinal obstruction	5
Acute reaction following gastroenterostomy (duodenal obstruction?)	2
Lobar pneumonia	2
Pernicious anemia	1
Acute pancreatitis with diabetes	1
Diabetes mellitus	1
Anaphylaxis (in experimental animals)	

The chemical findings in the blood and urine with the occurrence of melanuria can be illustrated by the chart of one case. The results here are typical of all the cases reported in this paper. This one is selected because recovery took place, and all stages of the relation can be traced. The patient developed a hemolytic streptococcus infection of the peritoneum following cholecystectomy. Subsequent to the peritoneal infection there was an obstruction in the ileum which necessitated an enterostomy. During this period the urine contained a large amount of melanin. The patient recovered.

Five months later she was again admitted with an obstruction of two days' duration. There was marked vomiting, abdominal pain and distention, and rapid pulse. Under non-surgical treatment the toxic symptoms all disappeared and the blood and urine returned to normal. On admission the blood chlorides were 300 mgms., and the urea nitrogen 31 mgs. per 100 c.c. blood. The urine showed only 0.01 per cent chlorides (as sodium chloride). The nitrogen of the urine was 1.8 per cent, and a large amount of melanin was present. As the chloride content rose and the nitrogen content fell to normal the melanin disappeared entirely from the urine. Simultaneously the blood chemistry returned to normal. These findings are shown in Table II and Chart I.

TABLE II.

Typical relation of chlorides and non-protein nitrogen of blood and urine to melanuria (case of intestinal obstruction).

Blood (mg. per 100 c.c.)	Sodium chloride	300			380					520	
	Urea nitrogen	31			49					10	
Urine (per cent)	Sodium chloride	0.01	0.01	0.03	0.06	0.02	0.08	0.20	0.27	0.45	0.62	0.53
	Nitrogen	1.8	1.6	1.3	1.4	1.2	1.2	0.8	1.1	0.3	0.4	0.4
	Melanin	++++	++++	+++	+++	+++	++	++	0	0	0	0
Day of disease		1	2	3	4	5	6	7	8	9	10	11

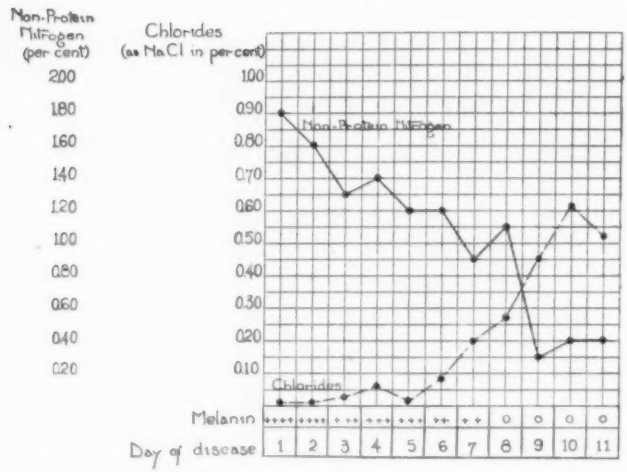

Chart showing the relation of the nitrogen and chloride content of the urine to the melanin content.

Discussion

Melanin was precipitated from the urine in all the cases here reported, so that its presence is unquestioned. In cases heretofore reported as occurring in the absence of melanotic tumor the method of identification used, in some cases at least, leaves some doubt as to whether melanin were really present. It has been pointed out that melanin should not be considered as present unless it can be actually precipitated out from the urine. Tests for alkaptone bodies, and for substances of the indican group which might be confused with melanin, were negative. There was no reason to suspect a melanotic tumor in any case, and such a possibility was actually ruled out by autopsy in two instances.

All these patients showed certain fundamental chemical features in common. During the period of melanuria there was uniformly a low blood and urine chloride, the nitrogen excretion was always high and usually there was an elevation of the nonprotein nitrogen of the blood well above normal. Such conditions give rise to a specific type of intoxication in seemingly widely unrelated clinical conditions. Optimal conditions are provided for the breaking down of protein in such a manner as to provide a large amount of the substances containing the oxyphenyl group from which melanin is formed. Conditions are evidently also favorable for the action of tyrosinase.

It is quite evident from the findings that the belief of many that melanuria is diagnostic of melanotic tumor is without foundation. The presence of melanin in the urine of a patient with manifest tumor of unknown type does not mean necessarily that the tumor is a melanotic sarcoma.

The great value of the demonstration of melanin in the urine in the absence of melanotic tumor would seem to be that it indicates a certain type of protein destruction. This abnormal protein destruction seemingly arises subsequent to and as a result of the depletion below a certain level of the available supply of sodium chloride. The possibility that autolysis may be initiated as a result of the sodium chloride depletion and thus explain the protein destruction has been discussed.

The presence of melanin in the urine in widely different clinical conditions is of interest, since it links together, chemically, clinical syndromes of different etiologic origin. It is probable that with further study it will be found in the urine during other diseases or conditions.

Summary and Conclusions

Melanins are complex protein substances formed by the action of tyrosinase on degradation products of protein containing the oxyphenyl group.

Normally, melanin is formed in the body through the activity of specialized cells, such as those of the choroid.

Under abnormal conditions melanin may be formed in the presence of an excessive breakdown of protein when the conditions for ferment action are favorable.

Twelve cases of melanuria are reported, all of which occurred in the absence of melanotic tumor.

All cases exhibited similar chemical changes within the body. There was always an increased protein destruction subsequent to a fall in blood chlorides.

It is evident that the presence of melanin in the urine is not necessarily indicative of a melanotic tumor.

The occurrence of melanin in the urine in the absence of a melanotic tumor is characteristic of a specific type of protein destruction which may occur in widely different clinical conditions.

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NOTES ON NEW BOOKS

Protein Therapy and Non-Specific Resistance. By WILLIAM F. PETERSON, M. D. *(New York, The MacMillan Company, 1922.)*

Dr. Peterson's treatise on protein therapy is a volume of two hundred and fifty-six pages. The contents are divided into eighteen chapters, an appendix, and a complete bibliography of fifty pages. The first seven chapters contain innumerable brief abstracts from the literature. They deal with the so-called non-specific protein agents and the non-specific reactions which occur after the injection of proteins. The theories of the reaction are also cited from the literature, but no attempt to criticise any of these theories is made by the author. The reading of these seven chapters is quite laborious, chiefly because of the great amount of repetition which naturally results from a compilation of abstracts, and for that reason the work will be of interest to very few clinicians.

The remainder of the book, eleven chapters, is a summary, together with a discussion of clinical results obtained by protein therapy. Innumerable diseases are taken up in detail—arthritis, typhoid fever, gonorrhœa and its complications, practically all of the infectious diseases, asthma, spirochetal and protozoan infections, diseases of the skin and eyes, and diseases of the central nervous system. The clinical observations are likewise abstracts from the literature.

As a reference book it has considerable value. The bibliography is excellent. The clinical reports are quite interesting, but they impress one as exaggerations of the true value of protein therapy. The writer deserves great credit for the enormous volume of work required to compile this treatise.

L. N. G.

A Text-Book of Pharmacology and Therapeutics. By E. POULSSON, Professor of Pharmacology, University of Christiania. 25 shillings. *(London, Wm. Heinemann, 1923.)*

This is one of the most popular text-books on pharmacology in Germany translated under the supervision of W. E. Dixon of Cambridge, himself the author of one of the most readable smaller English manuals on pharmacology. The German title "Pharmakologie" is loosely rendered as a "Text-book of Pharmacology and Therapeutics." This is perhaps better than the original German title, inasmuch as it emphasizes one of the most valuable features of the book, namely, that it is a treatise on pharmacology for students and the busy practitioner with especial regard to its applications in practice. In other words, it is a text-book on pharmaco-therapy as well as the science of pharmacology. The description of the action of drugs given here does not differ materially from that in other text-books on pharmacology except in certain of the explanations and hypotheses which serve to show something of the personality of the author and afford an insight into the Continental atmosphere. Thus, for instance, the discussion of camphor receives a rather undue importance, because that drug is extensively used on the Continent in heart conditions, whereas, in England and America, clinicians have found it to be of little value. The classification of the drugs in this book follows the old and somewhat illogical lines of Schmiedeberg's pioneer work on the subject. The first section deals with organic remedies acting as absorbents, the second section deals with organic substances acting locally, the third discusses salts of light metals, alkalis, acids, halogens, etc., oxidizers, etc., the fourth deals with heavy metals, and two smaller sections treat of ferments and antitoxins, etc. This arrangement is very much like that of the most popular text-book on pharmacology, that of Arthur R. Cushny. Poulsson's treatise, however, is more readable; it omits a great many purely pharmacological details which are of interest only to the specialist and lays stress on the relationship of pharmacology to applied therapeutics as an outstanding feature of the book.

Descriptions of various drugs are concise but are sufficiently complete for the student and general practitioner. A sentence or two in connection with all of the more important drugs gives a brief historical summary. The illustrations are not too numerous but are well selected and just enough chemistry

and chemical formulæ are incorporated into the work to give the reader an idea of the more important chemical-pharmacodynamic relationships without burdening his mind with unnecessary detail. Some of the sections are of exceptional excellence: Among these may be mentioned the chapter on cocaine, a field in which the author has done pioneer work. Again, the chapter on opium, antipyretics, purine derivatives and purgatives is exceptionally well written. The discussion of digitalis bodies which the author speaks of as the "digitoxin group" steers along conservative lines, so that the author, while adhering to the classical pharmacological conceptions on the subject, gives due regard to the more purely clinical contributions by recent investigators.

This text-book is very much up to date. There is a discussion of some of the newer drugs and new uses of older ones. The employment of quinidin in auricular fibrillation is described, and poisoning by wood alcohol is discussed in the light of recent experiences. There is a section on benzyl benzoate; the newer methods of treating syphilis with bismuth, etc., are mentioned and the final paragraph in the book makes a brief mention of insulin. As is inevitable in a translation, a few misprints or errors have crept in, as for instance on page 74 where the term benzoyl-isoquinolin is written in place of benzyl-isoquinolin. To the specialist in pharmacology the lack of a bibliography is a deplorable feature. Altogether, however, the book can be highly recommended to the medical student and the general practitioner.

D. I. M.

Clinical Surgical Diagnosis for Students and Practitioners. By F. de QUERVAIN. 3rd English edition. $14.00. *(New York, Wm. Wood & Co., 1921.)*

The translator's preface to the third English edition makes one rather hopeful that the character of the book may have been changed to a certain extent. But this hope is not to be realized. The most striking and interesting change in this newest edition is the number of illustrations which have been added to what was already a most unusual collection. Though published quite recently, comparatively little attention is given to the newer and undoubtedly more accurate forms of diagnostic methods, the lack being most marked in the matter of the various laboratory tests. In a book, written as it is for "students and general practitioners," one would expect to find diagnosis arrived at more through the logical consideration of various possibilities and the final conclusions reached by a process of elimination. But the author's great clinical experience makes much that is not so clear to his reader obvious to himself and in consequence the orderly type of differential diagnosis suffers. This rapid method of reaching conclusions certainly makes fascinating reading, but is of doubtful value as a teaching method. The arrangement of the material is also confusing. Though in a general way disease is considered from a regional standpoint, the individual conditions are classified in a quite arbitrary manner which makes constant reference to the index necessary. Rather frequent repetition does not add greatly to the clarity of the discussions. In Chapter VII the author epitomizes his volume in summing up his discussion of the surgery of exophthalmos, "We shall here, therefore, only collate a few points which have been treated disconnectedly, and supply sundry omissions."

The extensive chapter on surgical conditions of the neck is probably the best, and its interest is greatly added to by a series of splendid illustrations and case records. Apparently every tumor of the neck can be definitely diagnosed before operation—something rather far from the experience of most surgeons. As would be expected, the author devotes considerable space to diseases of the thyroid, but makes no mention of the Goetsch test, while referring to blood examinations and estimation of coagulation time as a "fashionable diagnostic method." His classification of lymph glandular tumors is very poor, but here again his discussion of clinical cases tends to make up for the more academic deficiency.

The diagnosis of thoracic conditions is quite confused, and disappointingly enough no practitioner could attempt an accurate diagnosis of cancer of the breast after reading the fairly large section devoted to it.

The approach to acute abdominal conditions is made with general peritonitis as the starting point, this being followed considerably later by sections on acute appendicitis and acute gastric and biliary conditions. The sequence here hardly represents the diagnostic aim of the general practitioner. There are too many excellent books available on diseases of bones and joints to warrant the use of three hundred pages in so general a volume as this.

Though undoubtedly interesting reading, the matter being presented in a most arresting style, we cannot help but feel that the book would have achieved its purpose equally well had illustrations and case records, in which the author is so rich, constituted the entire volume.

W. H.

THE JOHNS HOPKINS HOSPITAL BULLETIN

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